ON MYCOBOSCUS

Design Strategies & Epistemology of a Novel Sustainable Craft

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On Mycoboscus

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Design Strategies and Epistemology of a Novel Sustainable Craft.

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Abstract

With the advent of mycelium-based composites, we see the emergence of a novel material class. In excess of a technical description, this is a craft in budding. I call it that of mycoboscus.
Originating in the XIXth century use of solid-state fermentation for the transport of fungal strains intercontinentally, mycobosci emerged out of a logistical need like other fermentation-based crafts. Looking to implement the novel mycoboscus craft as sustainable is a unique opportunity that needs an early defining of the technique as frugal and vernacular to become an effective icon of fair and post-extractive dwelling.

To understand what the craft of mycoboscus comprises and establish a technical baseline, the thesis work first consists in the presentation of a biochemical and mechanical description of the system. This was done with a particular focus on the review and analysis of wood-decay fungi enzymatic activities.
Life-cycle and risks assessment studies were then screened to elaborate recommendations about the conditions for the sustainable cultivation and use of mycobosci. Given the ubiquitous capacity of chlamydospore synthesis in the fungal species that are considered, and the sensibility of the mycoboscus life-cycle assessment to the organic nature of the substrates, a scenario of local production for local distribution is recommen-

ded to limit environmental risks and increase the sustainability of the cultivation activity by resorting to by-products. This model constrains and weakens the technical system of mycoboscus by making it dependent on supplies of varying quality, thus leading to consider design strategies to address this systematically.
Three principal design strategies have been identified from this work. The supplementation strategy aims at chemically selecting or modifying the substrate to affect the enzyme activities, or the phenotypic expression of mycelia, for instance. Composition strategies consist of aggregates selection in the form of particles or fibres, for a diversity of shapes and strengths. Composition can also be achieved by designing composites with more than two phases to modify their behaviour under mechanical stress. The densification strategy can be executed by dense packing, cold or hot pressing.
The principles laid out for enacting supplementation show potential for adapting cultivation recipes to local opportunistic sources. While the densification strategy adds to the embedded energy of mycobosci and was not studied further for that reason, composition strategies were developed to increase the specific strength and stiffness of the composites efficiently. Novel layouts were tested experimentally in compression and flexion with arming, jacketing and failure initiators. The experimental results display a net performance increase with some designs, which corroborates the hypothesis that composition can be a sustainable and efficient design strategy, and useful for mechanical failure risk mitigation. The effect of particle sizes on compressive and tensile load response has also been studied, and a novel use for the specific matrix modulus metric is proposed for evaluating rapidly the size effect on fermentation respiration and binding quality.

It has only been, and yet already, about fifteen years since the first industrial development for mycobosci started. Whilst proven mechan-

ically competitive to EPS and XPS materials and more sustainable, the patent-protected industrialisation of mycobosci has not contributed to significantly reduce the use of these polluting solutions yet. The effect of the industrialisation process on the actual sustainability of mycobosci was studied through an integrative review. This use diffusion strategy is demonstrated to enhance the previously identified environmental risks that the technique presents, and shields knowledge from the public and academics. Industrialisation is shown to perpetuate the use of colonising and objectivising instruments that are detrimental to enacting sustainability, and hinder communities' capacity for auto-determination. This indicates a need for the conceptualisation of an alternative use diffusion strategy.

Knowledge production and maintenance is identified as a central interest for supporting these two entangled objectives of ecological and social sustainability. Based on the review of the mode of existence of vernacular architectures and terroir as eco-social designs, an alternative diffusion strategy is devised, and the concept of design terroir is formulated. This mode of existence is shown to favour situatedness and social individuation through a craft-based epistemology. It also exhibits a capacity for the democratic scrutiny of, and intervention upon existing technological infrastructures should they turn unsustainable, with the development and retention of communities' collective knowledge.

Epistemological hypotheses were formulated on the basis of this review work, and tested in the context of architectural education. This study resulted in an enhanced tacit knowledge appropriation in students through craft activities. This presents enticing opportunities for the potential contribution to eco-social development of designs that follow principles of design terroir formulated in this thesis.

With contributions in scientific description of mycobosci, technical development of sustainable material design strategies, and in relational

design theory, this thesis aims to support the emergence of a critical design research and practice of technology to foster the development of eco-social sustainability.

Abstrakt

Med fremkomsten af svampebaserede kompositter ser vi fremkomsten af en ny klasse af materialer. Ud over en teknisk beskrivelse er dette et håndværk i spirende. Jeg kalder det af mycoboscus.
Med oprindelse i det 19. århundrede brug af en solid-state fermentering, der blev brugt til at transportere svampestammer interkontinentalt, mycobosci opstod ud af et logistisk behov ligesom andre fermenteringsbaserede håndværk. At søge at implementere det nye mycoboscus-håndværk som bæredygtigt er en unik mulighed, der kræver en tidlig definition af teknologien som sparsommelig og sproglig for virkelig at blive et effektivt ikon for post-ekstraktive bolig.

For at forstå, hvad håndværket mycoboscus består af og etablere en teknisk baseline, består afhandlingsarbejdet først i præsentation af en biokemisk og mekanisk beskrivelse af systemet. Dette blev gjort med et særligt fokus på gennemgang og analyse af trænedbrydningssvampe enzymatiske aktiviteter.
Livscyklus- og risikovurderingsundersøgelser blev derefter screenet for at udarbejde anbefalinger om betingelserne for bæredygtig dyrkning og brug af mycobosci. I betragtning af den allestedsnærværende kapacitet af chlamydosporesyntese i de svampearter, der tages i betragtning, og følsomheden af mycoboscus livscyklusvurderingen over for sub-

straternes organiske natur, anbefales et scenarie med lokal produktion til lokal distribution for at begrænse miljørisici og øge bæredygtigheden af dyrkningsaktiviteten ved at ty til biprodukter. Denne model begrænser og svækker det tekniske system af mycoboscus ved at gøre det afhængigt af forsyninger af varierende kvalitet, hvilket fører til at overveje designstrategier for at løse dette systematisk.
Tre hoveddesignstrategier er blevet identificeret ud fra dette arbejde. Supplementeringsstrategien sigter mod kemisk at udvælge eller modificere substratet for at påvirke enzymaktiviteterne eller den fænotypiske ekspression af mycelier, for eksempel. Sammensætningsstrategier består af aggregatudvælgelse i form af partikler eller fibre, for en mangfoldighed af former og styrker. Sammensætning kan også opnås ved at designe kompositter med mere end to faser for at ændre deres adfærd under mekanisk stress. Tætningsstrategien kan udføres ved tæt pakning, kold- eller varmpresning.
Principperne for implementering af supplementeringsstrategien viser potentiale for at tilpasse dyrkningsopskrifter til lokale opportunistiske kilder. Mens fortætningsstrategien føjer til mycobosci indlejrede energi og ikke blev undersøgt yderligere af den grund, blev der udviklet sammensætningsstrategier for at øge den specifikke styrke og stivhed af kompositterne effektivt. Nye layouts blev testet eksperimentelt i kompression og fleksion med armering, jacketing og fejlinitiatorer. De eksperimentelle resultater viser en nettoydelsesforøgelse med nogle designs, hvilket bekræfter hypotesen om, at sammensætning kan være en bæredygtig og effektiv designstrategi og nyttig til at reducere risikoen for mekaniske fejl. Effekten af partikelstørrelser på kompressions- og trækbelastningsrespons er også blevet undersøgt, og en ny anvendelse af den specifikke matrixmodul-metrik er foreslået til hurtigt at evaluere størrelseseffekten på fermenteringsrespiration og bindingskvalitet.

Det er kun, og alligevel allerede, omkring femten år siden den første

industrielle udvikling for mycobosci startede. Selvom den har vist sig at være mekanisk konkurrencedygtig i forhold til EPS- og XPS-materialer og mere bæredygtig, har den patentbeskyttede industrialisering af mycobosci endnu ikke bidraget til at reducere brugen af disse forurenende løsninger markant. Effekten af industrialiseringsprocessen på den faktiske bæredygtighed af mycobosci blev undersøgt gennem en integrerende gennemgang. Denne brugsdiffusionsstrategi er demonstreret for at øge de tidligere identificerede miljørisici, som teknikken præsenterer, og beskytter viden fra offentligheden og akademikere. Industrialisering har vist sig at fastholde brugen af koloniserende og objektiverende instrumenter, der er skadelige for gennemførelsen af bæredygtighed, og hindrer lokalsamfunds kapacitet til selvbestemmelse. Dette indikerer et behov for konceptualiseringen af en alternativ brugsdiffusionsstrategi.
Videnproduktion og -vedligeholdelse er identificeret som en central interesse for at understøtte disse sammenfiltrede to mål om økologisk og social bæredygtighed. På baggrund af gennemgangen af folkelige arkitekturers og terroirs eksistensmåde som øko-sociale designs udtænkes en alternativ spredningsstrategi, og konceptet design terroir formuleres. Denne eksistensmåde er vist at favorisere situerethed og social individuation gennem en håndværksbaseret epistemologi. Det udviser også en kapacitet til demokratisk kontrol af og indgreb i eksisterende teknologiske infrastrukturer, hvis de skulle blive uholdbare, med udvikling og fastholdelse af lokalsamfunds kollektive viden.
Epistemologiske hypoteser blev formuleret på baggrund af dette reviewarbejde, og testet i sammenhæng med arkitektuddannelsen. Denne undersøgelse resulterede i en øget tavs videnstilegnelse hos eleverne gennem håndværksaktiviteter. Dette giver lokkende muligheder for det potentielle bidrag til øko-social udvikling af designs, der følger principperne for design terroir formuleret i dette afhandling.

Med bidrag i videnskabelig beskrivelse af mycobosci, teknisk udvik-

ling af bæredygtige materialedesignstrategier og i relationel designteori, sigter denne afhandling på at støtte fremkomsten af en kritisk designforskning og -praktik af teknologi for at fremme udviklingen af øko-social bæredygtighed.

Résumé

Avec l'avènement des composites à base de mycélium, nous assistons à l'émergence d'une nouvelle classe de matériaux. Au-delà d'une simple description technique, il s'agit d'un artisanat en plein essor. J'appelle celui-ci l'artisanat du mycoboscus.
Dérivés de l'utilisation de la fermentation en état solide pour le transport de souches fongiques à l'échelle intercontinentale au XIX$^{\text{ème}}$ siècle, les mycobosci sont apparus par nécessité logistique, à l'instar d'autres artisanats utilisant des principes de fermentation. Vouloir mettre à l'œuvre cet artisanat novateur du mycoboscus de manière soutenable représente une opportunité unique qui nécessite une définition précoce de cette technique comme frugale et vernaculaire afin qu'elle puisse devenir une icône de pratiques équitables et post-extractives.

Pour comprendre ce en quoi consiste l'artisanat du mycoboscus et établir une base technique, le travail de thèse consiste d'abord en la présentation d'une description biochimique et mécanique du système. Celle-ci a été réalisée en mettant particulièrement l'accent sur la revue et l'analyse des activités enzymatiques des champignons ayant la capacité de décomposer le bois.
Des analyses de cycle de vie et une évaluation des risques ont ensuite été examinées afin d'élaborer des recommandations sur les conditions de

culture et d'utilisation soutenable des mycobosci. Étant donné la capacité omniprésente de synthèse de chlamydospores chez les espèces fongiques considérées, ainsi que la sensibilité de l'analyse de cycle de vie du mycoboscus à la nature organique des substrats, un scénario de production locale pour une distribution locale est recommandé afin de limiter les risques environnementaux et accroître la soutenabilité de l'activité de culture en recourant de façon opportuniste à l'utilisation de déchets. Ce modèle contraint et affaiblit le système technique du mycoboscus en le rendant dépendant de l'utilisation de matières premières de qualité variable, ce qui conduit à envisager des stratégies de conception pour y remédier de manière systématique. Trois stratégies de conception principales ont été identifiées à partir de ce travail de revue. La stratégie de supplémentation consiste à sélectionner ou modifier chimiquement le substrat pour influencer les activités enzymatiques ou l'expression phénotypique du mycélium, par exemple. Les stratégies de composition consistent à sélectionner des agrégats sous forme de particules ou de fibres, avec une diversité de formes et de caractéristiques mécaniques. La composition peut également être réalisée en concevant des composites avec plus de deux phases pour modifier leur comportement sous charge. La stratégie de densification peut être réalisée par une préparation dense, un pressage à froid ou à chaud.

Les principes énoncés pour mettre en œuvre la supplémentation indiquent un potentiel pour l'adaptation de recettes de culture à des matières premières locales et utilisées de manière opportuniste. Puisque que la stratégie de densification contribue à augmenter l'énergie grise des mycobosci et n'a pas été étudiée plus avant pour cette raison, des stratégies de composition ont été développées avec pour objectif d'augmenter la résistance spécifique et la rigidité des composites avec efficience. De nouveaux designs de composition ont été testés expérimentalement en compression et en flexion, avec des renforcements, des chemisages et

des initiateurs de défaillance mécanique. Les résultats expérimentaux montrent une augmentation nette des performances avec certaines conceptions, ce qui corrobore l'hypothèse selon laquelle la composition peut être une stratégie de conception soutenable et efficiente, et utile pour atténuer les risques de défaillance mécanique liés à l'utilisation de substrats de qualité variable. L'effet de la taille des particules sur la réponse à la charge en compression et en traction a également été étudié, et une nouvelle utilisation du module d'élasticité normalisé de la matrice est proposée pour évaluer rapidement l'effet de la taille d'agrégat sur la respiration de la fermentation, et la qualité de la force de liaison entre agrégats.

Seulement quinze ans environ se sont écoulés depuis le début du premier développement industriel des mycobosci. Bien que ceux-ci se soient avérés compétitifs sur le plan mécanique par rapport aux matériaux EPS et XPS, et plus durables que ceux-ci, l'industrialisation sous brevets des mycobosci n'a pas encore contribué de manière significative à réduire l'utilisation de ces solutions polluantes. L'effet du processus d'industrialisation sur la durabilité réelle des mycobosci a été étudié ici à travers une revue intégrative. Cette stratégie de diffusion des usages des mycobosci démontre d'une augmentation des risques environnementaux précédemment identifiés et inhérents à cette technique, et occulte les connaissances techniques du public et des universitaires. Il est démontré que l'industrialisation perpétue l'utilisation d'instruments de colonisation et d'objectivation préjudiciables à la mise en œuvre de la soutenabilité, et entrave la capacité des communautés à s'autodéterminer. Ceci indique la nécessité de conceptualiser une stratégie alternative de diffusion des usages.

La production et la maintenance des connaissances sont identifiées comme centrales pour soutenir ces deux objectifs liés de soutenabilité écologique et de justice sociale. Sur la base de l'examen du mode

d'existence d'architectures vernaculaires et de la qualité de terroir en tant que conceptions éco-sociales, une stratégie alternative de diffusion est élaborée sous la notion de design de terroir. Ce mode d'existence favorise le situationisme et l'individuation sociale à travers une épistémologie basée sur l'artisanat. Il présente également une augmentation du potentiel à examiner et intervenir démocratiquement sur les infrastructures technologiques existantes si elles deviennent non soutenables, en favorisant le développement et la conservation de connaissances collectives au sein des communautés.
Des hypothèses épistémologiques ont été formulées sur la base de ce travail de revue, et ont été testées dans un contexte d'éducation architecturale. Cette étude a conduit à démontrer une appropriation accrue des connaissances tacites par les étudiant·e·s au travers d'activités artisanales. Ces résultats présentent des perspectives d'opportunités importantes pour la contribution au développement éco-social de designs qui suivent les principes du design de terroir formulés dans cette thèse.

Au travers de contributions concernant la description scientifique du système du mycoboscus, le développement technique de stratégies de conception durables, et à la théorie relationnelle du design, cette thèse vise à soutenir l'émergence d'une recherche en design et d'une pratique critique vis à vis de la technologie pour favoriser le développement d'une soutenabilité éco-sociale.

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The welcoming of fungi in my workspace did shake up considerably many ideas I had before. For instance, the idea that food should not be played with. With Chris Stewart and the team at Empirical I learnt that it *should* be played with, but only if this reduces spill. I am grateful to Chris for the enriching opportunity we had, and have, to close the loop.

The shared curiosity and openness is almost a method in itself, I learnt, gathering research interests and opening new areas of investigation. I thank Josh Evans for our wide ranging discussions that contribute to nourishing this curiosity.

I am thankful to the thr34d5 team, for the enthusing design discussions and practice we entertain, which reminds that there is a strong interest for the development of fair eco-social designs.

This work has been deeply inspired by Alpine culture and the ways some of its inhabitants co-become with places. This reflects a passion and *uncommon* that seeded a long time ago with foraging and wandering in steep mountainous paths. For this, and their renewed encouragement, inspiration and trust, I am thankful to Sylvie and Marc.

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Summary

Introduction

Cheese making and ripening practices have been critical to human development. From the practical need to store the nutritive proteins of milk to the crafting of the finest flavour profiles that make us belong, this ubiquitous interest spans civilisations, deserts, plains and mountains. Rich in its many forms, from the Lebanese *darfiyeh* to Nigerian *tchoukou* by way of the French *salers*, each tradition reflects not only its geography and climate but the organisation of the social life around it. Large cooked pressed cheeses from the French Alps for instance, reflect an organisation in co-operatives, where farmers gathered efforts to facilitate storage and distribution from areas seeing constrained logistics. In contrast, wetter and creamy cheeses such as *camembert*, originating from the plains, could be easily and quickly distributed, thus allowing for an adaptation of the craft (Delfosse 2007).

Just as cheeses, mycelium-based composites[1] originally came to exist as the result of a logistical need. Since the late XIXth century, solid-state fermentation (SSF) has been used by mycologists in Europe and the United-States to transport fungal strains between continents, on straw or other plant materials (Fig.1; Duggar 1905).

[1]Mycelium is the vegetative part of filamentous fungi. Mushrooms are the fruiting bodies of mycelia capable of synthesising such spore-bearing structures.

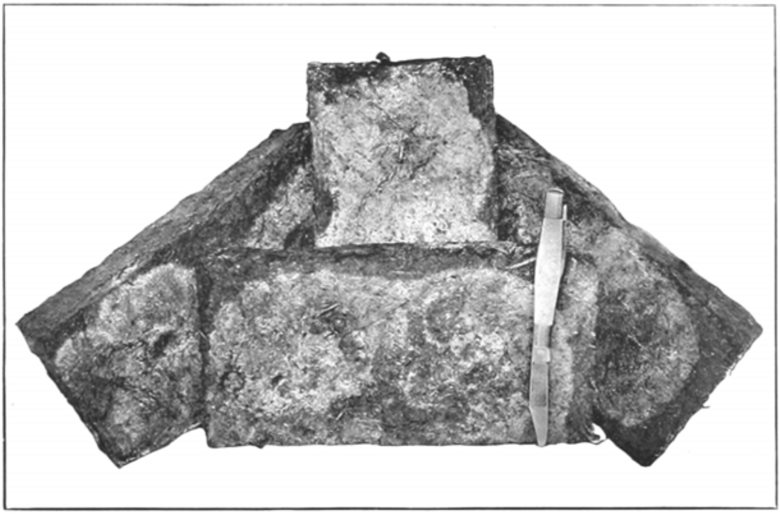

FIG. 2.—GOOD ("WELL-RUN") MUSHROOM SPAWN, BRICK FORM.

Figure 1: Mycelium spawn illustrated. Reprinted from Duggar 1905.

It is only recently that the capacity of wood-rot mycelia to bind wood debris has emerged as an independent interest. This is especially illustrated with artistic works from the United-States, which formed a ground for a global democratisation of the technique that started in the early two-thousands. A typical mycelium-based composite brick is illustrated in Fig.2. The cultivation method consists in having a moist cellulose-based material be colonised by a wood-decay fungus to a desired stage, and dry it to biological inactivation. The grown object, made of cellulose-based aggregates and a mycelium, is a composite material similar to wood-base fibre and particle panel materials.

Although this craft is novel, it is important to note that some of the fungal species in use already benefit from a rich cultural history. The species that was used throughout the present research work, *G. lucidum*, is also called *reishi* or *lingzhi* (靈芝). Its fruiting body has been widely used in Asian traditional medicine and is still a praised natural medicine to date. It is commonly referred to as *the fungus of immortality*, and its long-stipped fruiting bodies, or representations thereof, have been a symbol of long life, good health, good luck and prosperity in China. The earliest reported representation of the fruiting body as a *Ganoderma* sceptre—or *ruyi* (如意)—accompanies that of Emperor Xuan of Chen (陳文帝) and dates from the late VIIth century (Fig.3; Blanchette 2020). This history may influence cultivation practices and situated public reception of these fermented materials, and a designer might be careful to the histories of the fungal species they use as it taps into uncommons (section 4.2.6).

Mycelium-based composites represent an emerging, under-explored technology in a wide societal and cultural context—under conditions of the affordable democratisation of its craft knowledge. This is as if the development of leather work or wood-working were to be initiated nowadays, with an accompanying cultural capacity, and a perceived sustainable potential as these composites are made of a cellulose-based sub-

Figure 2: 1km geographical footprint mycoboscus brick (2022).

Figure 3: Detail from The Thirteen Emperors with a white ruyi. Yan Liben (second half of the VIIth century). Denman Waldo Ross Collection.

strate and mycelium. Furthermore, their cultivation can be as affordable as using foraged plant-material and fungal species, and rainwater.
This novel craft, I call it that of *mycoboscus* (plural *mycobosci*). Formed of the roots *múkēs* (μύκης, mushroom) and *boscus* (*bois*, wood, bush, woodland, to grow), mycoboscus defines the productions issued from the situated art of working with wood-rot fungi. The choice was made to use the suffix *boscus* rather than *lignum* (firewood; from Proto-Indo-European *leǵ-no-m*, that which is collected) to reflect the desirable geographical situating of the practice, formulated in the conclusions of this thesis. The terms of *mycelium-based composites*, or *mycelium materials*, do not do justice to the rich and transformative culture that the craft may support. Functionalist semantics would be reductionist as per the creative and cultural capacity lying in the practice, although useful in the context of material design to be rationally descriptive.

The development of mycoboscus cultivation was greatly influenced by the rich context of professional and amateur mycologists having flourished in the past 50 years, particularly in the United-States (Steinhardt 2018). Based on open science, invested amateur scientific curiosity and Do-It-Yourself methodologies, the contemporary mycobosci interest has been and is still promoted from third places on, and infuses in academia and industry from the margins. This grassroot thriving has been leading to a great promotion of mycological principles, and has fostered a wide social interest for the fungal kingdom. The artist Philip Ross[2] first started to experiment confidentially in the nineteen-nineties with the technique of mycoboscus in art production (P. Ross 2016), and with the growing popularity of his artworks and increase in grassroot projects visibility, commercial interests about mycobosci spun off. In 2007, the first commercial venture for composites was established (Ecovative Design, LLC).

[2] Philip Ross is now a co-founder and the chief technology officer of the company MycoWorks.

This founding along with the growing interest for Ross' artworks between 2009 and 2015 initiated a world-wide democratisation of the craft principles, specific to mycobosci. A wave of patents were filed from this moment on, with 13 patents assigned to Ecovative as of 2019 across mycobosci and pure fungal mycelium applications (Cerimi *et al.* 2019). A protective development leveraging industrial property seems relevant to support the business stability of a company centred on a technology with a particularly low barrier to entrance. This strategy about mycobosci displays an ambiguity with regards to the amateur mycologists network development having permitted this industrial establishment and millennia-old human fermentation practices based on peer-to-peer exchanges (Baumgarthuber 2021). While facilitating funding acquisition for one actor, this business development strategy impedes any potentially emerging actor, including those willing to implement this craft to speed up the adoption of patterns of sustainable production and consumption. It might also influence the actual sustainability of the products. This, as is well known, is occurring exactly when our societies need an urgent, humble and efficient change from industrial capitalism, to be sustainable and resilient in light of the current environmental collapse (Dhakal *et al.* 2022).

In this context of craft novelty, amateur practices and industrialisation, this thesis focuses on the investigation of the conditions for the sustainable cultivation and use of mycobosci:

> Under which conditions could mycobosci cultivation and use diffusion be sustainable?

To answer this question, design strategies for mycobosci are identified and qualified as of their relevance for sustainable practices; design strategies that allow for local and sustainable designs are evaluated; the effects that industrialisation may have upon the environmental risks mycobosci present are described, and an alternative use diffusion strategy is elaborated for enhancing the sustainability of mycobosci.

About fifteen years since the wider democratisation of the craft, its design space remains scarcely studied. While studies are being published now about mycobosci compostability, or about the mechanical behaviour of the composite system, recent life-cycle assessment studies indicate that the perceived sustainability of the cultivated materials is to be reconsidered. Additionally, a recent study has identified health and ecological risks of the cultivation of mycobosci, as per the spread of particular spores of ecologically competitive fungal species, and the potential release of volatile organic compounds during cultivation and use, for example. While the craft principles have been democratised, and the fields of fungal ecological studies and enzymatic biofuels production have advanced significantly in the recent years, a systemic description of the system for mycobosci design and cultivation has yet to be proposed from the perspective of enzymatic activities and their modification. The chapter II of this thesis proposes a semi-systematic review of the health and ecological risks, and focuses particularly on enzymatic activities and their modification, to identify risks and opportunities for cultivating sustainable mycobosci. A description of mechanical principles for these composites is also proposed to support the identification of opportunities, and three principal design strategies for mycobosci are identified. The findings from this review are used to outline an environmentally preferable scenario for mycobosci cultivation that conforms to locality for sourcing and distribution. This is to improve the sustainability of the system, and prevent environmental risks tied to the spread of ecologically competitive fungal species. While this scenario highlights the opportunity and desirability of vernacular, opportunistic byproducts upcycling, it also introduces a technological fragility. This fragility lies in the recommended use of opportunistic supplies, which will present varying qualities.

Based on the design strategies identified in chapter II, macro composition is studied to palliate a potential lack of consistency in principal substrate sourcing. Composition technical hypotheses are formulated in chapter III and tested experimentally. Scarcely investigated in the mycobosci state-of-the-art, aggregate size effect is also qualified mechanically. Moreover, the specific matrix modulus metric is introduced as an affordable method for qualifying the effectiveness of the fermentation, and is applied on qualifying aggregate size effect on aerobic respiration. Principles laid out in the chapter II, such as the impact of lignin nature and fungal species on the SSF, are quantified experimentally. Composition strategies are demonstrated to have a significant effect on mechanical performances, besides particular typologies. As a result, the significant strategies can be used to mitigate the risks tied to the opportunistic sourcing of substrates. The experimental results are presented comparatively to the state-of-the-art at the end of the chapter, and the distribution of species and principal substrate uses is mapped against mechanical performances.

Chapter IV is interested in identifying the effects that the patenting and industrial expansion has on the risks identified in chapter II. An integrative literature review is conducted with a focus on the principal dynamics of an industrial development from the perspective of scalability. As it is demonstrated that this uses diffusion strategy enhances greatly the ecological risks that mycobosci present, it is also shown to be detrimental to social resilience by hoarding and hiding knowledge for commodification[3].

As example of knowledge retention from a principal industry actor, significant gaps in materials and methods have been identified during the establishment of the current thesis in studies where this actor was a stakeholder (indicated by the presence of company owners as co-authors

[3]Commodification is the conceptualisation of material or immaterial things such as products, services, animals or ideas into objects of an economic value for trade (Appadurai 1994).

of studies). Non-disclosed fungal species are used in mechanical studies in Ziegler *et al.* 2016; L. Jiang *et al.* 2017; W. Sun *et al.* 2019; Ongpeng *et al.* 2020; L. Jiang *et al.* 2016; a non-disclosed substrate is used in L. Jiang *et al.* 2017; and non-disclosed species and substrate were used for informing a mechanical modelling in Islam *et al.* 2017. This is not an exhaustive report of the hindering of public knowledge and the process of verifiable, reproducible science by this company, but only concerns the studies reviewed during this research. The yearly data points contribution in the state-of-the-art is presented in Fig.4 with indication of the years having seen this retention happen. A data point is a unit defined in this thesis as a distinct mycoboscus design, i.e. a composite design defined by specific materials and methods.

This industrialisation of mycobosci is shown to both hamper the possibility of sustainable practices in chapter IV, and of knowledge dissemination for citizens. This may lead to a potential naturalisation[4] of polluting activities, holding up their potential investigation and redirection. In order to identify an alternative diffusion strategy, the recommendations from chapter II and the findings contextualising chapter IV about the effects of industrialisation are used. From this, the production and maintenance of knowledge within a community is identified as a central interest to support sustainability. Geographically and socially situated designs are then reviewed to identify leverages for community technological agency in terroir and vernacular design contexts. These cases are analysed, and recommendations are formulated for a sustainable eco-social[5] development in a design terroir context. This alternative strategy reflects a different method of knowledge production and maintenance, and supports

[4] The naturalisation of technology is the process by which technologies and designs come to be phenomenologically experienced by a user, and the service they provide is naturalised as a cultural axiom in one's worldview.

[5] Eco-socialism is interested in supporting the development of a socially just and ecologically sustainable society (Huan 2010).

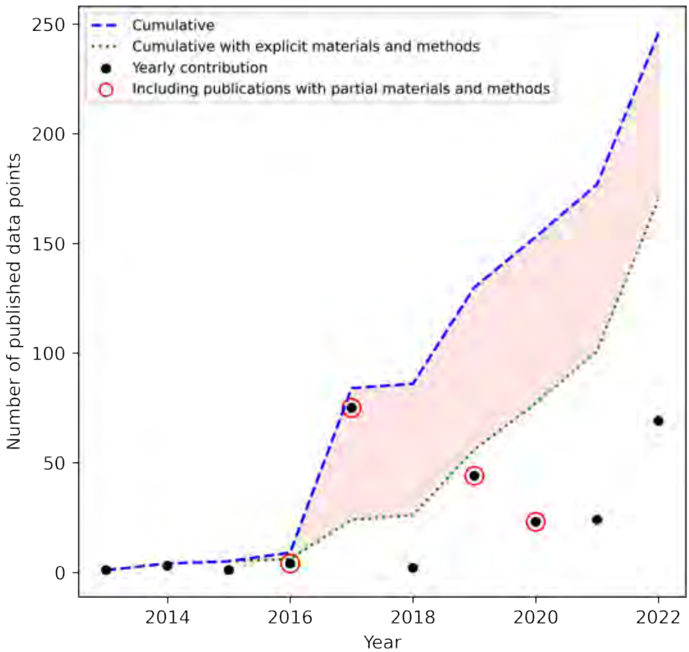

Figure 4: Published data points over time for mechanical studies considered in the state-of-the-art for this thesis. The complete publication list is presented in table 2.1. The years that include publications with partial materials and methods relate to non-disclosed fungal species in Ziegler *et al.* 2016; L. Jiang *et al.* 2017; W. Sun *et al.* 2019; Ongpeng *et al.* 2020; L. Jiang *et al.* 2016 and a non-disclosed principal substrate in L. Jiang *et al.* 2017.

a social individuation through design activities and use. This difference to an industrialised technological context is shown to be central for tying ecological sustainability and social equity and resilience. Finally, knowledge transmission hypotheses are formulated from this review work and tested in a pedagogical context of architectural education. These are corroborated, and show an important potential for leveraging the capacity of the craft of mycoboscus to influence a fair and just eco-social development.

This thesis elaborates on the social understanding of design and craft. The permeation of amateur mycologist works and philosophies into academic research may reflect in the emergence of research projects such as *Fungal Architectures* (European Union Future and Emergent Technologies H2020)—which was the framework for the development of this thesis. The experience of the craft of mycobosci cultivation invites deep sensations within the craftsperson and a careful and reciprocal engagement in the interaction with bizarre and delicate events (Fig.5). The sensible engagement with a fungal being made inert after having been blended, torn, structured, dried and revived appeals to a more-than-functional appreciation. This engagement draws a rift and permeates into academic practices from a corporeal origin. In Fungal Architectures, two traditional practices meet in research activities: kagome weaving and mycoboscus craft. Kagome is a triaxial weaving pattern that has been used in Japanese basketry for an indeterminate period. The motif exists as *kagome crest* (籠目) in Shinto, which comes in the form of a six and eight-pointed star, and is meant to represent the union of opposites like yin and yang[6]. The symbol seems to have been found in shrines dating back to the 5th century BC (Inocian *et al.* 2019). This assembly pattern has been used for its advantage in not necessitating screws or specific locking apparatuses as

[6]This pattern is also traditionally present in the Philippines under the name *bakat*. The origin of the pattern between Japanese and Filipino cultures has not been established (Inocian *et al.* 2019).

it is friction-based. Recently, rules for the placement of singularities in a triangulated base mesh for free-form kagome design have been instrumentalised in design tools to aid representation and exploration (Ayres *et al.* 2018). This makes kagome weaving a promising construction method for architectural applications, especially as the singularity rationalisation allows for the description of any free-form surfaces and can be produced using linear elements. Mycobosci on the other hand, although based on known principles of SSF, is a novel craft. As the Fungal Architectures research project aims to establish the ground for computing with living mycelia at architectural scale, it also has a third leg situating in the cybernetic tradition. The development of an ancient craft, the investigation of a novel one and the borrowing from recent reflections on information-based technologies makes for a rich context of development on the matter of *living architectures*. Balanced between circumstantial group works, technical investigations and artistic productions, the research hails for its integration into the larger society for its transformative potential by way of intimate sensible engagement with animal otherness in microbes and affordability of the kagome craft.

A selection of design works conducted during the Ph.D. period are presented in Fig.6, Fig.7, Fig.7, Fig.8 and Fig.9 to illustrate the aesthetics of mycobosci.

Figure 5: Ganoderma sublime. Students of the Liquid Modelling workshop (2020). Picture: own work (2020).

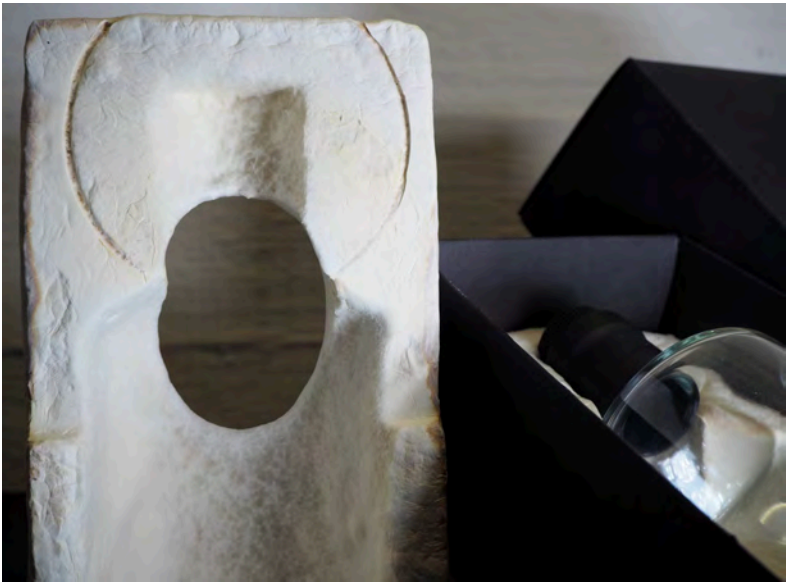

Figure 6: Ripened spirit case. Own work in collaboration with You-Wen Ji (2022). Picture: own work (2022).

Figure 7: Detail of ripened spirit case. Own work in collaboration with You-Wen Ji (2022). Picture: own work (2022).

Figure 8: Mycoboscus ripening (2022).

Figure 9: Mycoboscus prototype for the Kinoko Torii project, utilising upcycled materials for interaction design development. Own work in collaboration with thr34d5 (2023). Picture: own work (2023).

Chapter I – Methods

1.1 Design research

This study of mycobosci as a novel material class[7] and craft for sustainability ranges widely. From technical capabilities to people appreciation, the objects to investigate are quantitative and qualitative, shaped through engineering and design, experimental and philosophical. The research methods are therefore mixed and heterogeneous. They comprised: a semi-systematic scientific literature review; experiments based on the findings of this review; integrative literature review and a survey about a pedagogical experiment. Articulated by one principal research question, this work sets out to provide a comprehensive presentation of the findings for design researchers and practitioners interested in mycobosci and in designing for sustainability.
Tacit knowledge manipulation and production is core to design research. For instance, research-by-design is a method leveraging the design skills of the researcher to augment the tacit experience built across the invest-

[7] A material class is defined as a group of materials that share a coherent chemical makeup and atomic structure. Mycobosci, or mycelium-based composites, belong to the composites class. They represent a distinct material subclass within as they share chemical and mechanical qualities, along with process-specific methods (section 2.10, Fig.3.46, Fig.3.47).

igation and inform the research by subjective qualitative experiments—or prototyping. This is a method that acknowledges the usefulness of a research centred on sense-building. In fact, the importance of tacit knowledge in the designerly way of knowing can be connected to the identifying by Hubert Dreyfus and Stuart E. Dreyfus of it being a key component of intuition and sense-building in human cognition (Dreyfus *et al.* 2000)—although the identifying and conceptualising of tacit knowledge predates this work (Polanyi 1966). With the emergence of the field of design studies, Nigel Cross identified that a prominent quality of the iterative design method lies in its capacity to offer "*opportunities for development of a wide range of abilities in nonverbal thought and communication*" (Cross 1982). Looking to research as a systematic inquiry which results in novel, verifiable and communicable knowledge, the appreciation of Cross also points to the intrinsic effort that a designer would make to crystallise this knowledge into affordable results for an audience, and it has thus been thought to be particularly fit for research activities. Chris Rust also points to the usefulness of the documentation of artefact accumulation as a visual notebook of research traces in research-by-design for sense identification and building (Rust 2004). Considering the unique aesthetics of mycobosci, it was important to include more than textual accounts to try and render their specificity. To this end, pictures of experiments were included in this manuscript, and the use of graphical synthesis such as presented in Fig.2.17 also served as a tool to present a comprehensive scientific understanding towards dissemination for a general audience.

The research-by-design method is consequently a constructive approach, where the research arises from a design proposal, model or experiment, and results in generalising principle rules for the studied criteria (Hauberg 2011). The principal limit of this process is its intrinsic emergent positivism in that it invites to causation and generalisation from a situated exemplar. This aspect convokes the matter of the acknowledgement of a subjective perspective in the researcher, tending to auto-ethnographic methods that consist in using and reporting on self-reflection about anecdotal and personal experience in the research process (Marechal 2010). While the present research is not presented in a purely subjective form and was not defined from a solely constructive perspective, the representation of the sense-building activity is important to identify the relationship and coherence between findings of chapters II, III and IV.

Auto-ethnographic aspects of research-by-design were borrowed, in the valorising of craft somatic tacit knowledge and the conception and choice of use of the novel term *mycoboscus* throughout the manuscript, informed by the conclusions of the research. This semantic is meant to convey the essence of the neologism throughout the description of the specifics of the system, chapter by chapter, and to cultivate personal meaning in the reader by a snowballing effect. The choice of such a neologism as a white canvas is meant to convey controversies that may live asynchronously to the research timeline, controversies that are based on the research conclusions such as the recommended challenging of objectivising technical developments and industrialisation of mycobosci. *Mycoboscus* is employed as a conceptual tool borrowed from post-structuralist auto-ethnographic methodology heritage. While contributing to acknowledge the author position, this method also aims at "*remaking self and identity as a site for the negotiation of social, cultural and political dialogue*" (Marechal 2010). It results in a repeated evocation aiming to convey an intimate resonance, particularly by punctual linking to ubiquitous fermentation and cooking practices. This is a designed subjectivity in the presentation of the research. *Mycoboscus* locates (at) a singularity and was used as a platform to elaborate on design theory:

> Working by case is firstly to reason from singularities. Because the case is always and by definition what singularises

> in the middle of an ensemble of considered phenomena—either as a fact, when it presents itself objectively, or because it was chosen for its exemplary character. It is what produces a disruption, what distinguishes supremely within the sum of general laws supposedly capable of ruling the majority of possible occurrences.
> (author's translation; Davila 2019 p.24)

1.2 Scientific review

Looking to establish a substrate taxonomy for mycobosci, the perspective of the biological fundamental niche was used to conduct the review. The method that was adopted aimed at accounting a research area overview and identify both its development paths and informed future research directions. A semi-systematic literature review was chosen as the fittest method to achieve these goals, particularly to support a field meta-analysis (Snyder 2019). The articles that were considered are quantitative and peer-reviewed. The mycobosci state-of-the-art was reviewed systematically for reports of elasticity and strength in compression, flexion and tension. Other mechanical properties, or water absorption and thermal conductivity for instance, were not considered in the present work. Among the published works that are included in the state-of-the-art (listed in table 2.1) it is important to note that there is a strong heterogeneity in experimental methods and not all sources were showing statistically significant effects. Study specific discussions were thus not systematically conveyed in the research presented in this thesis, but the data has been analysed based on published materials and methods to render as completely as possible the status of the field. Mechanical reports without an explicit declaration of materials and methods utilised, were not included in the review, but for Ziegler *et al.* 2016; L. Jiang *et al.*

2017; W. Sun *et al.* 2019; Ongpeng *et al.* 2020; L. Jiang *et al.* 2016; Islam *et al.* 2017 as these data were found useful to draw a faithful picture of research dynamics.

The first aspect of the review consisted in establishing a state-of-the-art scientific understanding of the activity of the main actors of the mycoboscus system: wood-decay fungi. Looking to studies in plant material conversion for biofuels with ligninolytic enzymes, building and forest protection against fungal pathogens, saprotrophs and parasites, or enzymatic delignification for the paper industry for instance, the scientific review was conducted so to render most holistically the fungi degradation capabilities in relation to the substrate properties. Studies of fungal niche principles informed a systematic description of variables to consider for cultivating wood-decay fungi. With the perspective of use being closely tied to object mechanics, a review of activable properties of aggregate composites principles was also conducted. Using these two knowledge sets, driving principles informing design strategies for mycobosci have been identified and qualified, and outstanding values from the mycobosci state-of-the-art are discussed comparatively from this perspective. Meta-analysis was conducted on this state-of-the-art to help situate the advances of the field and the contribution of the present research. Given the aim in contributing to sustainable design, life-cycle and risk assessments for mycobosci were reviewed and concluded upon to feedback and funnel down recommended design use options. Based on a first set of conclusions from this work (section 2.12) experiments were designed to qualify the effect of composition strategies. The description of the materials & methods used for this are presented in section 3.2.

1.3 Relational design

The study into the effects of industrial development upon mitigating the environmental risks posed by mycobosci cultivation and use necessitated a work across several disciplines. From a designerly perspective—that of applied arts, i.e. culturally situated technique—the socio-technical entanglement of such a development entailed investigating the social and the technical effects at once. The method that was selected for this work is that of an integrative literature review. This review method is typically relevant for mature or emerging topics where the aim of the study is to critically analyse and synthesise the literature to enable new theoretical frameworks and perspectives to emerge (Torraco 2005). Rather than systematically reviewing a domain, this method focuses on the combination of perspectives from various domains allowing conclusions established from multiple end-points. This is particularly relevant to support a synthesising activity that characterises design activities. The principal research line for the integrative review is that of the notion of scalability and its effects on sustainability and public adoption in the context of mycobosci uses diffusion.

Based on the findings of this review work, knowledge transmission hypotheses have been established and tested in a pedagogical context. Teaching methods and afferent knowledge carriers were evaluated at the end of a workshop with a survey conducted across all participating students. The workshop focused on comparing the pedagogical efficacy for tacit knowledge acquisition of a digital mycelial growth simulation model, and craft activities. The method having been defined for the study presented in section 4.4 is elaborated in section 4.4.4.

1.4 Collaborative works acknowledgement

All of the research works reported here benefited from reviewing, funding acquisition and project management from Phil Ayres. Experimental works were designed under the supervision of Phil Ayres. The flexural characterisation experimental plan was executed by Claudia Colmo (section 3.2.5). Phil Ayres contributed to the execution of the compression testing (section 3.2.4). The integrative literature review on the aesthetics of *gross* was equally conducted together with Nadja Gaudillière-Jami (Rigobello and Gaudillière-Jami 2021). The study in pedagogical instruments effectiveness was equally conducted together with Nadja Gaudillière-Jami, who designed the survey of this study (section 4.4). The integrative literature review presented in chapter IV benefited from the supervision of Alexandre Monnin.

Chapter II – The Mycoboscus System

2.1 Introduction

To identify the influential parameters in mycobosci fermentation and the intrinsic characteristics that may inform design decisions, a description of the fermentation model is proposed from a semi-systematic literature review. This review was conducted with the principal aim of establishing a substrate taxonomy for mycobosci, taking as a principle the composite nature of the material and the intimate and microscopic relationship between mycelia and the substrate. Establishing a substrate taxonomy for mycobosci is an encyclopedic exercise. Similar to the development of an encyclopedia, what is critical is the identification of a cosmogonic classification system that will give a structure and sense to the content as research traces. In the tradition of Leibniz, the idea of an encyclopedia requires the evaluating of the relationship between the components of the system to identify their linking and principled description to satisfy a taxonomy that can serve as a base for existing knowledge, and a base for knowledge development (Eco 2011 p.78). As a linking focus, this study translates niche principles from ecological considerations, towards design inputs. The fundamental niche describes abiotic and biotic conditions under which a species can subsist and persist (Holt 2009). Illustrated by zone lines in decaying forest wood for instance, which represent a spatial method for some fungi to protect themselves against unfavourable environmental conditions or competitors in logs (Morris *et al.* 2021), the variations in genome across the surveyed species within the scope of this study display mutations and specifications that reflect ecological specifications, diets and behaviours.

After a brief introduction into the modes of sporulation and reproduction of filamentous fungi, and their nutritional needs, the description of the system is approached from the perspective of ligninolytic enzyme activity. Through reference to genome sequences and taxonomic efforts reported in the literature, the fungal species in use in the field of mycobosci can be qualified as per their enzyme expression capability and alternative species for potential future uses are identified. Learning from a recent human health and environmental risk assessment for mycobosci (van den Brandhof and Wösten 2022), a first set of design and use criteria are identified. This includes, for instance, the need to inactivate potential spores present in the mycelium before use and anticipate volatile organic compounds release. Plant-based substrates are then reviewed as per their cell configurations and cell wall composition, and the wetting capabilities and role of water in the system are presented. Having described the interactions between the wood-decay capability of fungi and the substrates through to wetting characteristics, various methods for regulating enzymatic activities and their effects, as reported in the literature, are presented. This first part of the study thus introduces a synthesis of the biochemistry of the SSF system from the angle of fungal enzymes and results in the identifying of design methods for modifying the substrate composition through, for instance, employing opportunistic raw materials.

Reviewing the mycobosci state-of-the-art and research efforts to describe pure mycelia mechanical behaviour, an understanding of influential parameters such as mycelium grown density is presented. While methods to modify the stiffness of mycelia can be derived from this review and previously introduced substrate composition strategies, the reported mechanical properties of four species display a wide range (tensile elastic moduli in 0.6 – 2727 MPa) and suggest that a variety of designs can also be cultivated. After a focus on substrate mechanical properties with particular interest in particulate materials and the influence of the decay activities on these, outstanding reports of mechanical characterisation are analysed from the mycobosci state-of-the-art. Three design strategies are identified from this scientific review: supplementation, densification and composition. The effect of these strategies on important mechanical properties are presented. In the midst of a rich number of influential variables on mycobosci mechanics and aesthetics, the introduction of these overarching strategies aims to support design decision making to compensate technical frailties that may subsist locally. These frailties may be necessary to maintain in the diffusion of the practice, in the sense that the mycoboscus life-cycle assessment (LCA) reviews point to a need to develop methods of production that are more energy efficient given the sensitivity of the system to incubation duration for instance, or the fact that it heavily relies on agricultural resources that have a significant impact on acidification, eutrophication and soil quality potential. Resorting to local byproduct upcycling may make the technical system more fragile given a potential lack of consistent supplies, while ensuring its sustainability. Tying this end to the recalling of the environmental risk presented by the ubiquity of active chlamydospores and their potential dispersal in any particular environment, composition strategies are identified as a possible frugal method to support a local supply to local use (*local-to-local*) scenario, while mitigating the risk in frailties.

2.2 Fungal biology background

2.2.1 Reproduction

Fungi are eukaryotes estimated to have originated on Earth 760 million to 1.06 billion years ago, while the plant kingdom is estimated to have evolved 820 million to 1.2 billion years ago (Money 2016a). The molecular clock dating method suggests that fungi have evolved to use wood as a nutrient since woody plants evolved (Watkinson 2016). The fungi kingdom is composed of fifteen phyla: Basidiomycota, Ascomycota, Glomeromycota, Entorrhizomycota, Blastocladiomycota, Chytridiomycota, Neocallimastigomycota, Monoblepharomycota, Olpidiomycota, Basidiobolomycota, Entomophthoromycota, Kickxellomycota, Calcarisporiellomycota, Mucoromycota and Mortierellomycota (Tedersoo *et al.* 2018). The species that are used in the making of mycobosci are largely basidiomycetes. The oldest fossil record of this phylum is 330 million years old and is distinct from its dikaryotic mycelium showing clamp connections—although the phylum is estimated to be older (Money 2016a). Basidiomycota represent about 30 000 known species covering yeasts to plant and human pathogens, and mushroom forming species (Deacon 2006 p.31). The basidium[8] is a characteristic feature of this phylum, in which meiosis occurs leading to sexual spore production. Basidiospores contain a single haploid[9] nucleus. They germinate and develop into monokaryotic mycelia[10]. A monokaryotic mycelium can produce oidia, asexually produced fungal spores that in contrast to conidia[11] are presumed not to constitute the main reproduction mode of the fungus. Oidia form as hyphae break up into cells and develop into spores. Oidia cannot survive in unfavourable

[8] Spore-bearing structure of basidiomycetes.
[9] Haploid describes a cell that contains a single set of chromosomes.
[10] Each monokaryotic hyphal cell contains one nuclear type.
[11] A conidium is an asexual spore produced by mitosis.

environmental conditions for long. Mycelia mating happens when two monokaryons of different compatibility groups fuse via plasmogamy[12], leading to a subsequent growth as a dikaryotic mycelium[13]. Dikaryotic mycelia can form fruiting bodies, where karyogamy will take place within each basidium—two nuclei fusing to form a diploid[14] nucleus. Monokaryotic basidiomycete mycelia, recognisable by a lack of clamp connections, generally do not produce fruiting bodies and the isolation of such strains can therefore find an interest for mycobosci cultivators[15]. Ascomycota can also be encountered in mycelium material practices, although rarely; the wood rot capabilities of taxonomic orders indicate a potential future research interest in this field. This phylum currently includes more than 33 000 species and is specific for its mating type involving an ascus, a cell in which plasmogamy happens (Money 2016a). The majority of ascomycetes produce asexual spores on conidiophores, mitospores. A number of ascomycete fungi are only known in their asexual forms (Deacon 2006 p.27). Chlamydospores, particularly resistant to unfavourable environmental conditions, result from an asexual reproduction. They result from a thickening of the cell wall of an hyphal compartment or can be formed on a chlamydoconidia apparatus (Money 2016b; Patakova 2013).

Basidiomycetes can be anamorphs and produce conidia, such as noticed in polypore white rot species *Ptychogaster rubescens* (Seifert and Gams 2001). This reproduction mode may involve the presence of spores within the mycelial structure. The presence of spore and fruiting body formation capacity, can represent a health and ecological risk as a respiratory health hazard and in the spreading of the species in the environment (van den

[12] Stage in fungi sexual reproduction in which the protoplasm of two parent cells fuse without nuclei fusion; it brings two haploid nuclei in the same cell.

[13] The dikaryotic mycelium contains the two nuclear types in fungal cells.

[14] Diploid describes a cell that contains two complete set of chromosomes.

[15] Monokaryotic fruiting bodies have been reported in, for example, *Mycoleptodonoides aitchisonii* (Riffiani *et al.* 2019). Monokaryotic and dikaryotic mycelia from species *Trametes versicolor* have been recognised to have similar decay activities (Hiscox *et al.* 2010).

Brandhof and Wösten 2022). The screening of species to use in mycoboscus cultivation should involve a risk assessment. The *Ganoderma* species have been reported to exhibit the capacity to produce chlamydospores in *G. colossus, G. oerstedii, G. rufoalbum, G. weberianum, G. multiplicatum, G. rothwellii, G. subamboinense, G. resinaceum, G. sessile* and *G. lucidum* (Moradali *et al.* 2007; Stalpers 2000). It was hypothesised that species of the *Ganodermataceae* family are regularly producing chlamydospores and show a higher resistance to elevated temperatures ($\geq$ 40 °C) (Loyd *et al.* 2019). *Trametes versicolor* strains have also been reported to produce chlamydospores (Bhatt *et al.* 2016), along with *Pleurotus subpalmatus* (Marryat 1908). Future research must investigate the inactivation conditions of chlamydospores for species in use in mycoboscus manufacturing to avoid introducing potentially pathogenic, parasitic or simply highly competitive species to an environment (Balaeș *et al.* 2023), which can influence local ecologies of decomposers (Fukasawa and Matsukura 2021). A study has been reporting fruiting body inhibition possibility in *Coprinopsis cinerea* and *Pleurotus djamor*, by targeting the glycogen synthase kinase-3 (GSK-3) regulatory protein using lithium chloride or CHIR 99021 (J. Chang *et al.* 2019). The growth medium profile has been shown to influence chlamydospore synthesis in the ascomycete *Trichoderma harzianum* Tr-92, and 2029 differentially expressed genes were identified under chlamydospore-producing conditions (Yuan *et al.* 2019). 2215 differentially expressed genes were identified in those conditions for the white rot *Phanerochaete chrysosporium* (L. Liu *et al.* 2020). Genetic modification has been studied to suppress chlamydospores synthesis capacity (Z.-B. Sun *et al.* 2019; Böttcher *et al.* 2016).

2.2.2 Nutrition

Fungi nutrients needed for growth consist of organic carbon (glucose, sucrose or starch are most common), nitrogen (ammonium, nitrate salts or organic amino acid), phosphorus, sulphur, potassium ions, calcium, magnesium and other compound traces (Watkinson 2016). Cellulose is the most abundant polymer form on Earth (Klemm *et al.* 2005) and can be decomposed by cellulase enzymes that cleave the chains to disaccharide cellobiose utilising cellobiosehydrolase (CBH) and endo-β-1,4-glucanase, which is cleaved by β-glucosidase to glucose monosaccharide by hydrolysis (section 2.3). Crystalline cellulose bundles present in plants are particularly resistant to hydrolysis. Nitrogen sources may be in the form of nitrate (NO_3^-) which is reduced to nitrite (NO_2^-) then to ammonium (NH_4^+) in the fungal cell (Watkinson 2016). When the carbon sources have been consumed, fungi can temporarily sustain their growth by assimilating CO_2 and use nitrate as a final electron acceptor in anaerobic respiration (Gunner and Alexander 1964). High concentrations of nitrates may be detrimental as they may trigger autolysis—the destruction of a cell under the action of its own enzymes. Ammonium is a preferred nitrogen source to nitrate (Watkinson 2016). Fungi have been shown to take up amino acids selectively, where core metabolic ones present in plants such as glutamate are readily assimilated compared to others like methionine (Watkinson 2016). Saprotroph fungi may also depolymerise proteins with proteases by hydrolysis and take up dissolved nutrients (Watkinson 2016). Phosphorus is central to fungal growth too. Phosphate (PO_4^{3-}) can be taken up from soil and water, and phosphate monoester and diester organic compounds along with inositol phosphate can be degraded by monoesterase, diesterase and phytase enzymes (Watkinson 2016). Sulphur is used by most fungi to synthesise proteins and glutathione peroxidase (Watkinson 2016; Schmalenberger *et al.* 2011; X. Yang *et al.* 2010). Potassium and magnesium are essential to fungal growth

too, such as lower amounts of iron, copper, calcium, manganese, zinc and molybdenum as cofactors of proteins (Watkinson 2016).
Fungi can produce antibiotics as secondary metabolites[16] to protect nutrient sources from bacterial competitors. *G. lucidum* has been reported to display an antibacterial activity (Hassan *et al.* 2019) potent to *Micrococcus luteus* (Yoon *et al.* 1994), *Escherichia coli*, *Salmonella typhimurium* and *Bacillus subtilis* (Sheena *et al.* 2003), and *Auricularia auricula* against *Staphylococcus aureus* (Iftekhar *et al.* 2011), for instance. This is mainly due to laccase expression capability and its antibacterial activity (Mathur *et al.* 2021).

2.3 Enzymatic activity in ligninolytic fungi

Pleurotus, *Ganoderma* and *Trametes* are the most frequently cited families in mycoboscus design. *Schizophyllum commune* is a less investigated species but finds a growing interest (Appels *et al.* 2020) especially because it is has a sequenced genome and is one of the few mushroom-forming fungi for which genes have been inactivated by homologous recombination (Ohm *et al.* 2010)[17]. This strategy has proved useful for genetic studies and functional genomics (Sawitzke *et al.* 2013). Gene inactivation articulates the principles of homologous recombination for replacing an existing autologous gene with a designed heterologous gene. Furthermore, the recombinant DNA constructs of *S. commune* has been reported to express in other mushroom-forming fungi, such as with *Pycnoporus cinnabarinus*, which supports this species importance for fungal biology (Ohm *et al.* 2010; Alves *et al.* 2004). A list of species having been reported for mechanical characterisation of mycobosci is presented in table 2.1—the referenced studies constitute the state-of-the-art of this thesis (see

[16] Activated by carbon or nitrogen depletion (J. Yang *et al.* 2017).
[17] The main content of the following of this text until section 2.10 was the topic of a research disseminated in Rigobello and Ayres 2023; appendix E.1

reported species to substrate combinations on Fig.3.49 and Fig.3.50). A list of species having been reported for other characterisations is presented in tables 2.2 and 2.3. The species listing was elaborated from previous review works (van den Brandhof and Wösten 2022).

Wood-rot fungi can be classified by their decay activity, which cover three main modes: white-rot (specified as selective delignification, and simultaneous rot), brown-rot, and soft-rot. The decay modes are illustrated in Fig.2.10. Across the most used species, all of them are white-rot, ligninolytic fungi. They have the enzymatic ability to decompose all three principal polymer classes found in wood: lignin, hemicellulose, and cellulose, but most often degrade primarily lignin and hemicellulose such as reported for *G. lucidum* (Chen *et al.* 2012). As such, they qualify as selective delignifiers, as opposed to simultaneous decomposers where all three polymer classes endure similar decay rates. Brown-rot fungi uses a two-step oxidative-enzymatic mechanism for the breakdown of cellulose and hemicellulose (Brischke and Alfredsen 2020). Their lignin degradation abilities remain under-investigated, and although brown-rot fungi is a polyphyletic group evolved from at least seven white-rot lineages (Floudas *et al.* 2012), they reportedly lack more than 60 % of the genes known to be involved in white rot. Brown-rot species degrade cellulose and hemicellulose at a higher rate than white-rot ones in laboratory isolation (Castaño *et al.* 2018). This decay mode is most often encountered in coniferous trees. Soft-rot fungi commonly share the ability to demethylate. This mode is associated with ascomycota and monokaryotic mycelia, and is distinctive for its tunnelling decay pattern through lignin layers to areas richer in cellulose and hemicellulose (Schwarze 2007). The environmental conditions for enzyme activation help define the species niche and their subsequent distribution. The principal wood-rot enzyme families are presented in Table 2.4.

Lignin linkages result from radical reactions (Sangha *et al.* 2012), in contrast to polysaccharide polymerisation resulting from hydrogen bonding (Diotallevi 2007 p.7). Lignin therefore does not involve hydrolytic enzymes and it cannot be used as a source of carbon or energy for most wood-rot fungi: its decomposition serves primarily for the fungus to gain access to hemicellulose, pectin, and cellulose compounds (Watkinson 2016). Lignin depolymerisation is mostly initiated by aromatic[18] ring oxidation by peroxidases (by use of H_2O_2 or R-OOH) (Gargulak *et al.* 2015; Hofrichter *et al.* 2010; Vanholme *et al.* 2010; Boerjan *et al.* 2003), while other aromatic compounds in the cell structure are mostly catabolised by monooxygenases and dioxygenases. Peroxidases are hemeproteins[19] (Wong 2009). LiP are known to oxidise phenolic aromatic substrates[20] along with various nonphenolic lignin compounds, and other organic compounds with a high redox potential. MnP leads to H_2O_2 oxidising Mn^{2+} to Mn^{3+}, an oxidising compound to monomeric phenols. Mn^{2+} chelates[21] permeate through wood cells and decay lignin selectively. These chelates cannot oxidise nonphenolic compounds, but radicals formed by Mn^{3+} oxidation can in turn oxidise benzyl alcohols and other diarylpropane structures (Kuan *et al.* 1993; Wong 2009). MnP can also peroxidise lipids (Watanabe *et al.* 2001). Versatile peroxidase (VP) are enzymes that are Mn^{2+} specific but can oxidise phenolic and nonphenolic compounds in its absence (as with LiP). They are involved in oxidation of high redox potential aromatic substrates, thus reducing Mn^{2+}-independent oxidation but not affecting Mn^{2+}-dependent one (Tinoco *et al.* 2007). Mn^{2+}-independent oxidation capabilities for phenols, small dye compounds, and amines has also been described for MnP (*Pr*MnP2, *Pr*MnP3;

[18] Aromaticity defines a cyclic and typically planar molecular structure. These molecules are more stable than single bonds and non-cyclic structures.

[19] Proteins that contain an iron cation and activate O_2 for oxidation or hydroxylation.

[20] Phenols are aromatic organic compounds consisting of a phenyl group ($-C_6H_5$) and an hydroxyl group (–OH).

[21] Chelates are organic compounds formed when an ion or molecule bonds with more than two donor atoms to a central metal atom.

Hofrichter *et al.* 2010), and it is predicted from sequencing that similar divergent MnP may be expressed in *Trametes* spp., *Pleurotus* and *Agaricus bisporus* among other species (Zhang, Dong *et al.* 2021; Lundell *et al.* 2010; Hofrichter *et al.* 2010). Hydrogen peroxide, necessary for LiP and MnP activities, is the product of laccase activity (Schlosser and Höfer 2002). A peroxidase family with wide substrate specificity—of which anthraquinones[22]—and active at pH of approximately 3 has been identified in fungi: dye-decolourising peroxidase (DyP; Kim and Shoda 1999). Laccases are a very versatile family of enzymes: they are expressed in nearly all fungi including ones without lignin degrading abilities (Baldrian 2006). They perform functions ranging from synthesis of melanin and other pigments to conidia, fruiting body formation, and, indeed, lignin decomposition (Eggert *et al.* 1996). These belong to a multicopper enzyme family and catalyse the removal of an electron from phenolic hydroxylic groups to form phenoxy radicals which polymerise via radical coupling. Ultimately, the reactions lead to the presence of oxidised quinones and couples oligomeric products. Laccases also have a wide substrate specificity and can act upon an extended range of phenols, diphenols, aminophenols, and aromatic compounds. When their catalytic reaction is accompanied by demethylation it may lead to ring cleavage (Venkatesagowda and Dekker 2021). Tying lignin decomposition to cellulose and hemicellulose decay, cellobiose dehydrogenase (CDH) has the ability to oxidise cellobiose, cellodextrins, and lactose (Zamocky *et al.* 2006). Hemicellulose degrading enzymes can have the ability to hydrolyse various polymers, such as illustrated by β-1,4-galactosidase that degrades xyloglucan, xylan, galactomannan, and pectin (Kubicek 2012 p.70). More specific xyloglucanases, xylanases, mannanases and pectinases can be found in all fungi to a various extent. Typically, pectin can be hydrolysed by polygalacturonidases, but also cleaved by β-elimination by pectin and

[22] Naturally occurring phenolic compounds, building block of many dyes.

pectate lyases (Jayani *et al.* 2005; Seyedarabi *et al.* 2010). Cellulose hydrolysis by fungal enzymes employs glycoside hydrolases (GH) for glycosidic bonds cleaving. Among GH families, cellobiohydrolase I (CBH I) is the major cellulase being produced in white and soft rot; it binds to crystalline cellulose and hydrolyses it into cellobiose. CBH II, found in all but brown-rot fungi, binds to amorphous cellulose and separates cellobiose from the non-reducing end of cellulose using acid catalysis of the *β*-1,4-glycosidic bond (Goodell 2020; Fleming and Pfaendtner 2013). Endoglucanases are a major enzyme group able to hydrolyse cellulose polymer from within the molecule rather than its end (Goodell 2020). The variety encountered in fungi refers to four GH families: GH5, GH7, GH12, and GH45. GH5 endoglucanase is a less studied family in the case of fungi. GH7 endoglucanase displays a similar mechanism to CBH I, with the difference that it can hydrolyse from the middle of cellulose molecules (Goodell 2020). GH12 endoglucanase lacks a carbohydrate-binding module (CBM), they are therefore unable to degrade crystalline cellulose but can hydrolyse it in its amorphous form (Sidar *et al.* 2020). GH45 endoglucanase benefits from a CBM (Sidar *et al.* 2020). *β*-glucosidases, produced by all fungi, belong to GH1 and GH3 families. GH1 *β*-glucosidases are able to cleave soluble *β*-linked oligosaccharides from up to nine glucose residues chains and aglycone-linked *β*-glucosides (Hrmova *et al.* 1998), and can be competitively inhibited by the products of CDH activity (Kubicek 2012 p.64). GH3 *β*-glucosidases can remove single glucosyl residues from the non-reducing ends of oligo- and polysaccharides such as *β*-D-glucans, *β*-1,3-D-glucans, *β*-1,4-D-glucans, to name a few (Hrmova *et al.* 1998). Lytic polysaccharide monooxygenases (LPMO) operating by oxidative cleavage have been shown to enhance cellulose degradation (Harris *et al.* 2010; Couturier *et al.* 2018).

Carbohydrate-active enzymes (CAZymes; Cantarel *et al.* 2009) have been classified in six principal families: Auxiliary Activities (AA), Glycoside Hydrolase (GH), Glycosyltransferase (GT), Polysaccharide Lyase (PL), Carbohydrate Esterase (CE), and Carbohydrate-Binding Module (CBM). The glycosyltransferase reaction involves the transfer of a monosaccharide from a nucleotide sugar[23] to a saccharide, protein, lipid, DNA or small molecule acceptor (Ünligil and Rini 2000; Horton 2008). They are involved in fungal cell wall synthesis, protein and lipids glycosylation (Klutts *et al.* 2006). Carbohydrate esterases have a wide substrate specificity and are involved in the degradation of acetylated hemicelluloses (Biely 2012). They release the acyl or alkyl groups attached by ester linkage to saccharides (Armendáriz-Ruiz *et al.* 2018).

The genome distributions of *T. versicolor*, *P. ostreatus* PC15, and *G. lucidum* G0119 are presented according to this nomenclature in table 2.5. *G. lucidum* benefits from the largest known CAZymes array with 565 genes having been assigned to these functions (Zhou *et al.* 2018; Chen *et al.* 2012). This species is a well known versatile fungus capable of selective delignification—this is confirmed experimentally in section 3.3. The actualisation of decay strategies in selective delignification capable fungi has been correlated to wood species, lignin contents, temperature, pH, and moisture content (Freschet *et al.* 2012; Parfitt *et al.* 2010; Schwarze, Engels *et al.* 2000; Adaskaveg *et al.* 1995; Blanchette 1995). Mycoboscus manufacturing protocols adjusting for enzyme activities regulation is presented in section 2.7.

2.4 Fungal species for mycobosci

The species that were reported and included in the present mycobosci state-of-the-art, are all basidiomycetes belonging to the class of agarico-

[23] Nucleotides are organic molecules forming the base of DNA and RNA. They are made of a nitrogenous base, a five-carbon sugar and a phosphate. Nucleotide sugars act as glycosyl donors in glycosylation reactions catalysed by glycosyltransferase.

Species	Order	Life-history strategy	Decay mode	Reference
Agaricus bisporus	Agaricales (B)	Saprotroph	Secondary decomposer	Tacer-Caba *et al.* 2020
Coprinus comatus	Agaricales (B)	Saprotroph	White rot	Houette *et al.* 2022
Ganoderma lucidum	Polyporales (B)	Saprotroph, parasitic	White rot	Rigobello and Ayres 2022; Rigobello, Colmo *et al.* 2022; Tacer-Caba *et al.* 2020; R. Liu *et al.* 2019; Silverman *et al.* 2020; R. Liu *et al.* 2020; Travaglini *et al.* 2013; Travaglini *et al.* 2014; Heisel *et al.* 2018
*Ganoderma resinaceum**	Polyporales (B)	Saprotroph, parasitic	White rot	Elsacker *et al.* 2021; Elsacker *et al.* 2019
Ganoderma resinaceum	Polyporales (B)	Saprotroph, parasitic	White rot	G. Angelova *et al.* 2021
Irpex lacteus	Polyporales (B)	Saprotroph	White rot	Z. Yang *et al.* 2017
Lentinus crinitus	Polyporales (B)	Saprotroph	White rot	Cesar *et al.* 2021
Lentinus velutinus	Polyporales (B)	Saprotroph	White rot	Bruscato *et al.* 2019
Pleurotus ostreatus	Agaricales (B)	Saprotroph	White rot	Appels *et al.* 2019; Tacer-Caba *et al.* 2020; Silverman *et al.* 2020; Sivaprasad *et al.* 2021; Mohd Nashiruddin *et al.* 2022; López Nava *et al.* 2016; Sağlam and Özgünler 2022; Alemu *et al.* 2022; Houette *et al.* 2022
Pleurotus albidus	Agaricales (B)	Saprotroph	White rot	Bruscato *et al.* 2019
Pleurotus citrinopileatus	Agaricales (B)	Saprotroph	White rot	Silverman *et al.* 2020
Pleurotus eryngii	Agaricales (B)	Saprotroph	White rot	Silverman *et al.* 2020
Pycnoporus sanguineus	Polyporales (B)	Saprotroph, parasitic	White rot	Bruscato *et al.* 2019
Trametes multicolor	Polyporales (B)	Saprotroph	White rot	Appels *et al.* 2019
Trametes versicolor	Polyporales (B)	Saprotroph, parasitic	White rot	Elsacker *et al.* 2021; Elsacker *et al.* 2019; Lelivelt *et al.* 2015
Trichoderma asperellum	Hypocreales (A)	Endophyte	–	Tacer-Caba *et al.* 2020
Non-disclosed	–	–	–	Ziegler *et al.* 2016; L. Jiang *et al.* 2017; W. Sun *et al.* 2019; Ongpeng *et al.* 2020

Table 2.1: Species in use in mechanical characterisation studies in flexion, tension or compression integrated to the state-of-the-art. A: ascomycetes, B: basidiomycetes. *The *G. resinaceum* species in use in these studies has been requalified as *G. lucidum* after sequencing.

Species	Order	Life-history strategy	Decay mode	Reference
Agrocybe aegerita	Agaricales (B)	Saprotroph	White rot	Attias *et al.* 2017
Allomyces arbusculus	Blastocladiales (BL)	Water saprotroph	–	Jones, Huynh *et al.* 2018
Bjerkandera adusta	Polyporales (B)	Saprotroph, plant pathogen	White rot	Appels *et al.* 2020
Botrytis cinerea	Helotiales (A)	Necrotroph	Grey mould	Jones, Huynh *et al.* 2018
Ceriporia lacerata	Polyporales (B)	Saprotroph, endophyte	White rot	Shao *et al.* 2016
Coprinellus micaceus	Agaricales (B)	Saprotroph	Wood rot	Appels *et al.* 2020
Coprinopsis cinerea	Agaricales (B)	Saprotroph	Litter-degrading	J. Chang *et al.* 2019
Deadaleopsis confragosa	Polyporales (B)	Parasitic	White rot	Bajwa *et al.* 2017; Appels *et al.* 2020
Flammulina velutipes	Agaricales (B)	Saprotroph	White rot	Tacer-Caba *et al.* 2020
Fomes fomentarius	Polyporales (B)	Saprotroph, parasitic	Simultaneous rot	Moser *et al.* 2017; Stelzer *et al.* 2021; Freidank-Pohl *et al.* 2022
Fomitopsis pinicola	Polyporales (B)	Saprotroph	Brown rot	Wimmers *et al.* 2019
Fusarium oxysporum	Hypocreales (A)	Saprotroph, endophyte, plant pathogen	White rot	Jones, Huynh *et al.* 2018
Ganoderma applanatum	Polyporales (B)	Saprotroph, parasitic	White rot	Vallas and Courard 2017
Ganoderma lucidum	Polyporales (B)	Saprotroph, parasitic	White rot	Jones, Huynh *et al.* 2018; Răut *et al.* 2021; Soh *et al.* 2021; R. Liu *et al.* 2020
Ganoderma resinaceum	Polyporales (B)	Saprotroph, parasitic	White rot	Bajwa *et al.* 2017
Ganoderma sessile	Polyporales (B)	Saprotroph, parasitic	White rot	Attias *et al.* 2020
Gloeophyllum odoratum	Gloeophyllales (B)	Saprotroph	Brown rot	Appels *et al.* 2020
Gloeophyllum sepiarium	Gloeophyllales (B)	Saprotroph	Brown rot	Wimmers *et al.* 2019
Hypsizygus ulmarius	Agaricales (B)	Saprotroph, parasitic	Brown rot	Jones, Huynh *et al.* 2018
Kuehneromyces mutabilis	Agaricales (B)	Saprotroph	White rot	Appels *et al.* 2020; Tacer-Caba *et al.* 2020
Laetiporus sulphureus	Polyporales (B)	Saprotroph, parasitic	Brown rot	Wimmers *et al.* 2019
Lentinula edodes	Agaricales (B)	Saprotroph	White rot	Tacer-Caba *et al.* 2020
Lichtheimia corymbifera	Mucorales (M)	Saprotroph, animal pathogen	–	Jones, Huynh *et al.* 2018
Megasporoporia minor	Polyporales (B)	Saprotroph	White rot	Xing *et al.* 2018
Mucor genevensis	Mucorales (M)	Saprotroph	–	Jones, Huynh *et al.* 2018
Oxyporus latemarginatus	Hymenochaetales (B)	Plant pathogen	White rot	Xing *et al.* 2018
Phaeolus schweinitzii	Polyporales (B)	Parasitic	Brown rot	Wimmers *et al.* 2019
Phytophthora cinnamomi	Peronosporales (O)	Plant pathogen	–	Jones, Huynh *et al.* 2018
Piptoporus betulinus	Polyporales (B)	Parasitic	Brown rot	Wimmers *et al.* 2019

Table 2.2: Species in use in other characterisation studies (1/2). A: ascomycetes, B: basidiomycetes, BL: blastocladiomycetes, M: mycoromycetes, O: oomycetes.

Species	Order	Life-history strategy	Decay mode	Reference
Pleurotus citrinopileatus	Agaricales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018
Pleurotus cornucopiae	Agaricales (B)	Saprotroph, parasitic	White rot	Jones, Huynh *et al.* 2018
Pleurotus djamor	Agaricales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018; J. Chang *et al.* 2019
Pleurotus eryngii	Agaricales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018; Teixeira *et al.* 2018
Pleurotus ostreatus	Agaricales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018; Appels *et al.* 2019; Wimmers *et al.* 2019; Attias *et al.* 2017; Silverman *et al.* 2020; Soh *et al.* 2021; He *et al.* 2014; Helberg *et al.* 2019; Kuribayashi *et al.* 2022; Teixeira *et al.* 2018; J. Zhang *et al.* 2019; J. Zhang *et al.* 2019
Pleurotus pulmonarius	Agaricales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018; Attias *et al.* 2017
Pleurotus salmoneostramineus	Agaricales (B)	Saprotroph	White rot	Attias *et al.* 2017
Polyporus arcularius	Polyporales (B)	Saprotroph	White rot	Wimmers *et al.* 2019
Polyporus brumalis	Polyporales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018; M. P. Jones *et al.* 2019
Pycnoporus sangineus	Polyporales (B)	Saprotroph, parasitic	White rot	Teixeira *et al.* 2018; Bruscato *et al.* 2019; Santos *et al.* 2021
Saksenaea vasiformis	Mucorales (M)	Animal pathogen, saprotroph	–	Jones, Huynh *et al.* 2018
Schizophyllum commune	Agaricales (B)	Saprotroph, parasitic	White rot	Appels *et al.* 2020
Stropharia rugosoannulata	Agaricales (B)	Saprotroph	White rot	Jones, Huynh *et al.* 2018
Trametes betulina	Polyporales (B)	Saprotroph	White rot	Appels *et al.* 2020
Trametes hirsuta	Polyporales (B)	Saprotroph, plant pathogen	White rot	Appels *et al.* 2020; Kuribayashi *et al.* 2022
Trametes multicolor	Polyporales (B)	Saprotroph	White rot	Appels *et al.* 2020
Trametes pubescens	Polyporales (B)	Saprotroph, plant pathogen	White rot	Wimmers *et al.* 2019
Trametes suaveolens	Polyporales (B)	Saprotroph	White rot	Wimmers *et al.* 2019
Trametes versicolor	Polyporales (B)	Saprotroph	White rot	Bajwa *et al.* 2017; Jones, Huynh *et al.* 2018; M. P. Jones *et al.* 2019; M. Jones *et al.* 2019; Jones, Bhat *et al.* 2018; Appels *et al.* 2020; Attias *et al.* 2020
Trichaptum abietinum	Hymenochaetales (B)	Saprotroph	White rot	Wimmers *et al.* 2019
Xylaria hypoxylon	Xylariales (A)	Saprotroph	White rot	Appels *et al.* 2020

Table 2.3: Species in use in other characterisation studies (2/2). A: ascomycetes, B: basidiomycetes, BL: blastocladiomycetes, M: mycoromycetes, O: oomycetes.

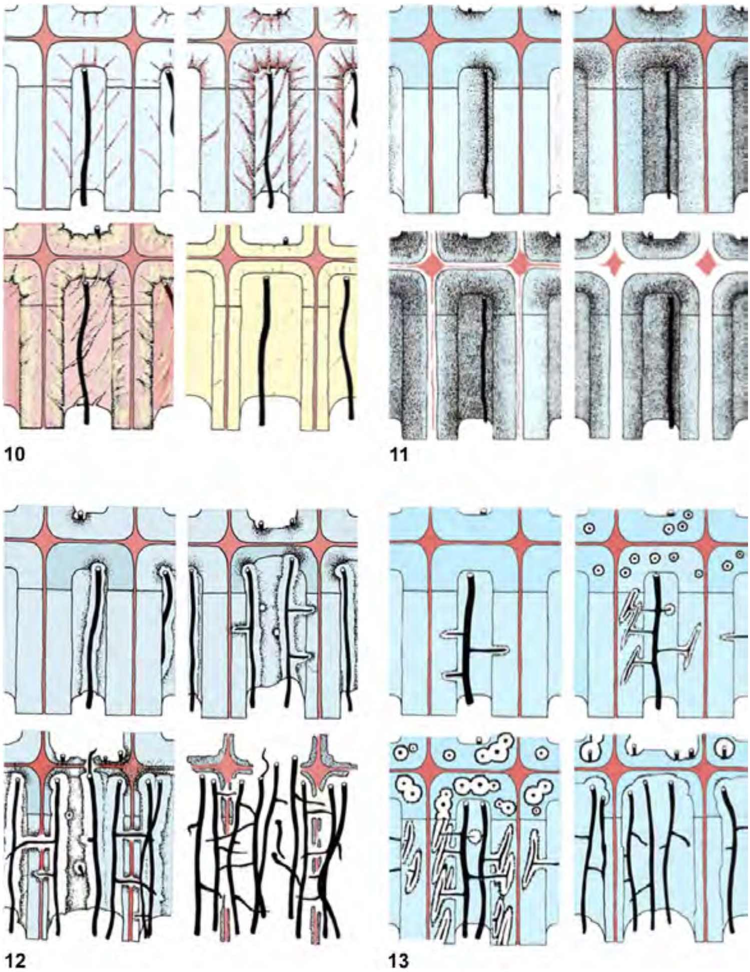

Figure 2.10: Schematic drawings showing micro-morphological features of different decay types: (10) brown rot by *Fomitopsis pinicola*; (11) selective delignification by *Heterobasidion annosum*; (12) simultaneous rot by *Fomes fomentarius*; (13) soft rot by *Kretzschmaria deusta*. Reprinted from Schwarze 2007.

Target polymer	Enzyme family	Reaction	Substrate	Temperature	pH
Lignin	Lignin peroxidase	Oxidation	Phenol, nonphenolic aromatics.	28 – 40 °C	3.0 – 4.5
	Manganese peroxidase	Oxidation	Manganese ions, phenol, small dye compounds, amines.	23 – 40 °C	5.0 – 7.0
	Versatile peroxidase	Oxidation	Manganese ions, phenol, nonphenolic aromatics.	35 °C	7.0
	Dye-oxidising peroxidase	Oxidation	High redox-potential dyes, nonphenolic lignin model compounds.	28 – 70 °C	3.0 – 7.0
	Laccase	Oxidation	Phenol, diphenol, aminophenol, aromatic compounds.	35 – 50 °C	4.0 – 5.0
Hemicellulose	Xyloglucanase	Hydrolysis	β-1,3-glucans, carboxymethyl cellulose, xyloglucan oligosaccharides.	37 – 75 °C	3.0 – 9.4
	Xylanase	Hydrolysis	glycosidic bonds in xylan backbone.	28 – 50 °C	3.0 – 9.0
	Mannan-degrading	Hydrolysis	Mannan, galactomannan.	45 – 92 °C	2.4 – 7.5
	Polygalacturonidase	Hydrolysis	Pectin.	–	–
	Pectin lyase	β-elimination	Heavily methyl-esterified.	50 °C	8.0
	Pectate lyase	β-elimination	Lower esterification and Ca^{2+}.	30 – 90 °C	5.0 – 11.5
	Cellobiose dehydrogenase	Oxidation	Cellobiose, cellodextrins, lactose.	60 °C	3.0 – 10.0
Cellulose	Cellobiohydrolase I	Hydrolysis	Crystalline cellulose.	40 – 80 °C	–
	Cellobiohydrolase II	Hydrolysis	Amorphous cellulose, β-xylan.	50 – 63 °C	4.0 – 6.0
	GH5 endoglucanase	Hydrolysis	Cellulose.	45 – 65 °C	4.5 – 7.0
	GH7 endoglucanase	Hydrolysis	Crystalline cellulose.	45 – 65 °C	4.5 – 7.0
	GH12 endoglucanase	Hydrolysis	Amorphous cellulose.	45 – 65 °C	4.5 – 7.0
	GH45 endoglucanase	Hydrolysis	Crystalline cellulose.	45 – 65 °C	7.0
	GH1 β-glucosidase	Hydrolysis	β-linked oligosaccharides, aglycone-linked β-glucosides.	25 – 50 °C	5.0 – 7.0
	GH3 β-glucosidase	Hydrolysis	β-glucan.	25 – 50 °C	5.0 – 7.0

Table 2.4: Extracellular enzymes in lignicolous fungi and their optimal activation temperature and medium pH. Adapted from Kubicek 2012; Zamocky *et al.* 2006; Tinoco *et al.* 2007; Gomes *et al.* 1992; Paramjeet *et al.* 2018; Vyas *et al.* 1994; F. Jiang *et al.* 2008; Ravichandran *et al.* 2021; Kwang-Soo *et al.* 2005; Rajhans *et al.* 2020; Desai *et al.* 2011; Rashmi and Siddalingamurthy 2018; Motta *et al.* 2013; Moreira and Filho 2008; Yadav *et al.* 2009; Wu *et al.* 2020; Cameron and Aust 2001; Nataraja *et al.* 2010; Heinzelman *et al.* 2009; Teixeira da Silva *et al.* 2016.

CAZy family	Enzymes	TV	PO	GL
AA	Laccase, LiP, MnP, VP, DyP, CDH, LPMO	89	114	96
CBM	GH5 – GH7 – GH45 endoglucanase, CBH I	47	82	33
GH	GH12 endoglucanase, β-glucosidase, xyloglucanase, xylanase, polygalacturonidase, mannan-degrading, CBH II	222	235	262
GT	Glycosyltransferases	85	67	72
PL	Pectin lyase, pectate lyase	9	23	11
CE	Carbohydrate esterases	19	28	91

Table 2.5: Genome distribution of CAZymes from selected species: *T. versicolor* (TV) (Sista Kameshwar and Qin 2018), *P. ostreatus* PC15 (PO) (Sista Kameshwar and Qin 2018), *G. lucidum* G0119 (GL) (Zhou *et al.* 2018).

mycetes (table 2.1). This class represents the only organisms capable of substantially degrading lignin (Floudas *et al.* 2012). *Trichoderma asperellum* is an exception as it is an ascomycete (Tacer-Caba *et al.* 2020) and is used to regulate pathogenic organisms in plant crops and lead to increased yields.

Future species to be evaluated for their use in mycoboscus cultivation can be initially identified from their genome by comparing their degradation capacities to the substrates at hand[24]. Higher gene reads for class II peroxidases (LiP, MnP, VP) allowing lignin degradation are identified from sequencing in model species *Phanerochaete chrysosporium* and *Auricu-*

[24] *Trichoderma reesei* is a cellulolytic species of industrial interest for cellulase production. Its CAZymes-related genes are not displaying higher reads than less effective cellulose degrading species, but its greater cellulolysis capacity is related to its secretion efficiency (Watkinson 2016). Species selection by CAZymes-encoding gene expression analysis is therefore a limited methodology.

laria delicata, *Fomitiporia mediterranea*, *Dichomitus squalens* and *Punctularia strigosozonata*, for instance (Floudas *et al.* 2012). These species are good candidates to expand the mycoboscus species repertoire. Moreover, *P. chrysosporium* has been previously reported to have a radial extension 2.6 – 9.2 times higher than that of a *G. lucidum* strain on substrates of manure, wood chips, potato dextrose yeast extract (PDYA) and yeast synthetic defined (YSD) (Perez *et al.* 2020). *Phlebia tremellosa* (Blanchette *et al.* 1988), or *Dichomitus squalens* that is a species preferring softwood, could also be of interest to practitioners (Daly *et al.* 2018; Blanchette *et al.* 1988). It can be noted that *S. commune* is considered a white rot species although it has a limited lignin decay capacity with no gene reads for class II peroxidases (Floudas *et al.* 2012).

The species are best selected for their absence of chlamydospore production capability to ensure product stability and reduce health hazards. Some species such as *I. lacteus* have been reported to cause pulmonary abscess in immuno-suppressed persons (Buzina *et al.* 2005). Saprotrophs can be plant pathogens, such as illustrated by white rot *Stereum hirsutum* that is found as pathogen in peach trees. *Xylaria* spp. and *Hypoxylon* spp. are wood decaying ascomycetes, but as for *T. asperellum* that was already reported in a mycoboscus study, their conidia formation capability may represent a health hazard. The selection of fungal species for mycoboscus cultivation and use should involve precaution. A recent study surveyed the human health and environmental risks of the practice (van den Brandhof and Wösten 2022). The study points to respiratory infections having been reported for *S. commune* (Chowdhary *et al.* 2013; Michel *et al.* 2016) which the authors relate to some agaricomycetes having the potential to become opportunistic human pathogens. They also point to the intrinsic risk related to the fact that species are selected for their higher colonisation rate and degradation potential which reflects their ecological competitiveness. The species therefore represent a risk to local environments, crops and orchard viability, and wooden structures (for *Ganoderma* spp. see: Vinjusha and Arun Kumar 2022; Lodha and Harsh 2010; Paterson 2007; Susanto *et al.* 2005; Sankaran *et al.* 2005, for others species: Gabriel and Švec 2017; M. Fischer 2006). Fungi may produce volatile compounds that repel or attract animals and insects depending on their cultivation conditions, and the widespread democratisation of their use could impact insect biodiversity and infestation risks (van den Brandhof and Wösten 2022; Bajwa *et al.* 2017; Korpi *et al.* 2009; Kabbaj *et al.* 2002).

Widespread species reproducing sexually have a higher genetic variability (Urbanelli *et al.* 2003). This may impact the reproducibility of mycoboscus studies and necessitate local protocol adaptations. To illustrate, it was reported that the degradation of birch samples by five strains of species *P. chrysosporium* resulted in a widely ranging lignin loss of 23 – 73 %, a 10 – 50 % glucose loss and a 28 – 58 % xylose loss for an overall weight reduction of 17 – 46 % (Blanchette *et al.* 1988). The difference in gene reads in species *G. lucidum* G0119, *P. ostreatus* PC15 and *T. versicolor* (table 2.5) also illustrates the different decay behaviours that may express phenotypically. As the CAZy AA family gathers enzymes being involved in a variety of functions, it would be unfit to comment on the reads at this level of clustering. The CBM family broadly targets crystalline cellulose, the GH family amorphous cellulose and xylans, CE hemicelluloses and the GT family is involved in cell wall synthesis. The higher number of gene reads in the GH and CE families in *G. lucidum* G0119 reflects a greater versatility in hemicelluloses degradation and a preference to amorphous cellulose—although it has high lignin degrading capability as previously reported. *P. ostreatus* PC15 seems more equipped to degrade lignin and oxidise its substrates with 114 gene reads, and its crystalline cellulose degradation capability is clearly higher than that of the two other species. *T. versicolor* high GT reads may point to a great diversity of functions involved in cell wall synthesis, which can influence its decay activity considering a potential influence on membrane porosity or mycelium colonisation patterns. In addition to its impact on reproducibility, aesthetics and performance, genetic variation and mutation present an environmental risk as a species can see a change from endophytic to pathogenic lifestyle with a single locus mutation (van den Brandhof and Wösten 2022; Saikkonen 2007).

Beyond life strategies of fungi, their strain specific genomes and life history can influence the specific decaying capabilities for a same cultivation protocol and substrate. The genetic selection and *in-vitro* gene regulation by media modifying over generations of sexual reproduction may lead to species gene expression modification and induce an evolution.

2.5 Lignocellulosic substrates

Lignin is the most effective barrier to fungal decay in lignocellulosic substrates as it prevents migration of low molecular weight diffusible agents (i.e. H_2O_2, veratryl alcohol, oxalate, Fe^{2+}, Fe^{3+}, Mn^{2+}, Mn^{3+}; Evans *et al.* 1994) for decomposing cellulose and hemicellulose. The nature of lignins is of considerable influence on their sensitivity to enzymatic activity. Lignins are polymers of phenylpropene units, they are composed of guaiacyl, syringyl and p-hydroxyphenyl units (Whetten and Sederoff 1995). Gymnospermous woods (conifers) are almost exclusively composed of guaiacyl monomers, while angiospermous trees (hardwood) contain approximately equal shares of guaiacyl and syringyl monomers (Whetten and Sederoff 1995). Similarly, different distributions of hemicellulose compounds are found in gymnospermous trees (where galactoglucomannan, glucomannan, and arabinoglucuronxylan are the principal ones) and in angiospermous trees (where arabinogalactan, xyloglucan, and various glucans are the principal compounds) (Schwarze 2007). Pectins are primarily made of β-1,4-D-galacturonase acid units and their methyl esters, interrupted locally by 1,2-linked L-rhamnose units (F. Green *et al.*

1996). The primary constituent of wood is cellulose, constituted by repeating glucose units joined by β-1,4-glucosidic linkages (Kadla and Gilbert 2000). Cellulose represents about 50 % of the wood cell wall, lignin 25 %, hemicellulose 20 to 25 %, and pectin 1 to 4 %. Hemp is often in use in mycoboscus practice. The lignocellulosic compounds of its tow, with the ratio cellulose:hemicellulose:lignin is close to 15:2:1, when its shives display a ratio closer to 2:1:1. The cultivation period and climate has been reported to induce differences in non-cellulosic composition (van der Werf *et al.* 1994).

The variety of wood structures and chemical compositions pairs with various decay strategies driven from the set of fungus CAZyme expression. These wood specifics can be understood from the evolutionary history of cell specialisation. Principle wood structures are presented in Fig.2.11. Wood cells can be made of various specialised cells: tracheids, xylem ray parenchyma, axial parenchyma, and vessels. Tracheids are found down to a first evolutionary stage of woody plants (trees) in gymnospermous woods, where they perform both water transport and structural stiffness. Gymnospermous woods typically lack vessels and rarely have axial parenchyma, their tracheids to xylem ray parenchyma ratio is most often in the vicinity of 9:1 (Schwarze 2007). They contain principally guaiacyl lignin, a product of coniferyl alcohol (Fengel and Wegener 2011). In angiospermous woods, water is transported by vessels while tracheids have a structural function. Another evolutionary stage of water transport system in trees can be exemplified by European beech (*Fagus sylvatica*) and birch (*Betula pendula*). Their xylem includes vessels along with tracheids, the former being a distinct location of guaiacyl lignin (Schwarze 2007). Tracheids and parenchyma contain a fraction of syringyl lignin spread across the cell wall layers, making this secondary evolutionary wood structure attractive for white-rot fungi (Schwarze 2007). A third evolutionary stage can be illustrated by European oak (*Quercus robur*), for which the xylem is ring-porous containing early and late wood vessels in bands within a thin-walled tracheids matrix. The strength of the wood comes from libriform fibres containing higher syringyl lignin, while tracheids have a higher guaiacyl content (Schwarze, Baum and Fink 2000a). A late evolutionary stage is exemplified by sycamore (*Acer pseudoplanatus*) and ash (*Fraxinus* spp.), where syringyl dominates (Schwarze, Baum and Fink 2000b). Here, vessels are dedicated to water transport, and are jacketed in living libriform fibres that are included amongst dead ones. Guaiacyl lignin has been reported to have a higher resistance to fungal decay (Schwarze 2007), which has been corroborated by phylogenetic and experimental study showing that catalytic tryptophan presence in more recently evolved peroxidases (located in LiP and VP families) was more efficient at oxidising angiospermous woods, while others performed better in gymnospermous woods (Ayuso-Fernández *et al.* 2019). Early stage parenchyma decomposition, and a resistance from libriform fibres to white-rot decay, has been confirmed experimentally (Schwarze 2007).

The structure of the wood cell wall composing the xylem, is composed of a middle lamella (ML), primary wall (P), and three layers of secondary wall (S1, S2, S3). They are represented with their fibre orientation and indicative thicknesses in Fig.2.12. The structuring middle lamella is composed of lignin, calcium, and pectic compounds that are amorphous. Crystalline cellulose is most prominent in younger wood, so it dominates the S3, and is found in reducing percentages moving towards the P. Inversely, hemicellulose can be found in greater extents in the P and in reducing shares towards the S3, while lignin is quite evenly spread across the cell wall, with higher concentrations in the ML (Schwarze 2007). The distribution of lignocellulosic compounds in woods is presented in Table 2.6, and an illustration of wood cell wall specification in plants is presented in Fig.2.13.

Fungal colonisation of wood happens through pit apertures or boreholes

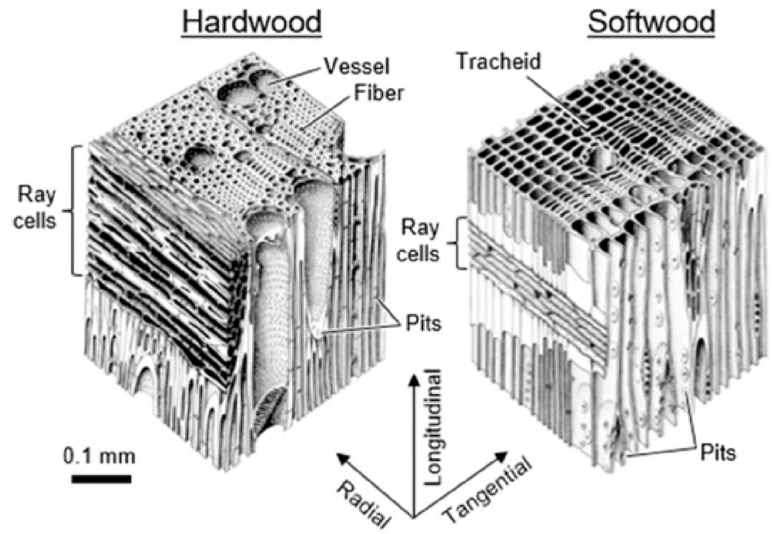

Figure 2.11: Principle structures of soft and hardwood. Reprinted from Jakes *et al.* 2019.

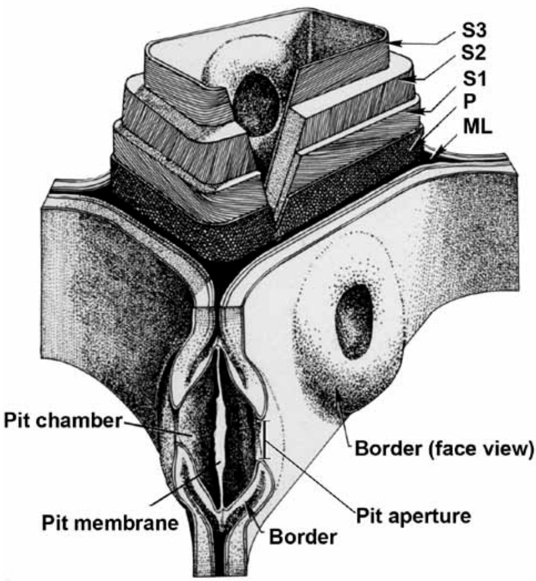

Figure 2.12: Wood cell wall structure: middle lamella (ML), primary wall (P), secondary wall (S1, S2, S3). Reprinted from Wiedenhoeft 2010.

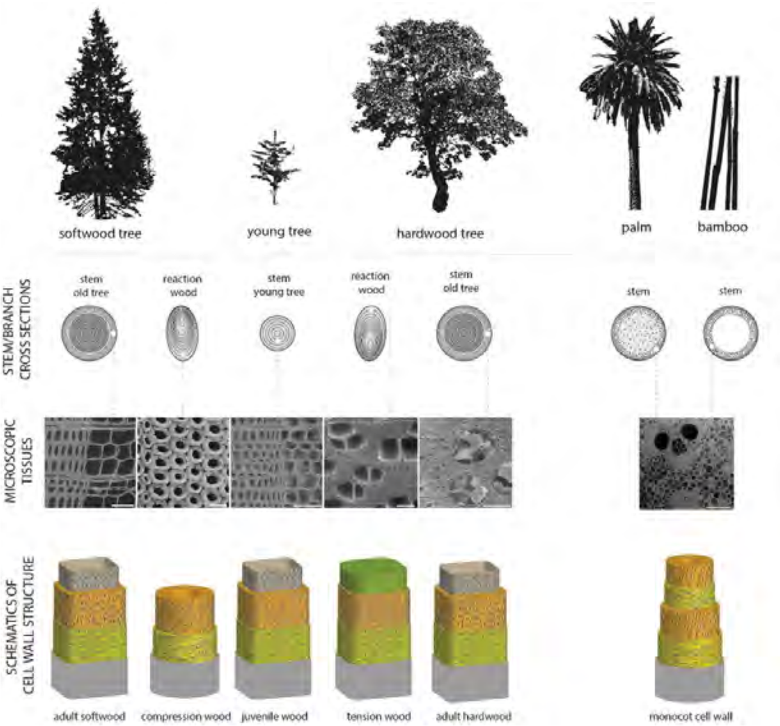

Figure 2.13: Structure of wood cells. Microscopic tissues from left to right: adult spruce wood (bar 50 µm), spruce compression wood (bar 20 µm), juvenile spruce wood (bar 50 µm), poplar tension wood (50 µm), adult oak wood (bar 200 µm), and vascular bundle of bamboo (bar 200 µm). Last row showing schematics of cell wall structures. Light gray indicates the compound middle lamella (0.5 – 1.5 µm) and primary cell wall (approximately 0.1 µm), yellow S1 layer with a thickness of 0.1 – 0.35 µm, orange S2 layer as 1 – 10 µm thick; inner light gray layer shows the 0.5 – 1.1 µm thin S3 layer; the green G-layer in tension wood fibres can fill the whole lumen and the cellulose fibrils are oriented along the longitudinal cell axis. Reprinted from Eder *et al.* 2021.

induced by enzymatic activity (Wilcox 1970). Although pectin contents are marginal in wood, pectinase expression is critical to decomposing the pit membrane (Fig.2.12), which also contains cellulose, and allow the fungus to proceed in colonising the wood (Schwarze and Landmesser 2000). Calcium is associated with pectin in wood; Ca^{2+} can combine with fungal oxalic acid and result in pectic acid which solubilises pectin (Peng *et al.* 2020; F. Green *et al.* 1996). Calcium, potassium, phosphorus and magnesium are the major elements associated with plant fibre. The mineral content (ash) in softwood is 0.02 – 1 wt% and 0 – 5 wt% in hardwood (Tsoumis 2009). Large concentrations of calcium oxalate and residues of MnO_2 accumulate during white-rot decay (Blanchette 1995). Wood can, moreover, contain a variety of compounds that may be toxic, mainly polyphenols or tannins in angiosperms and phenolic compounds in gymnosperms (Schwarze and Ferner 2003). Some fungi that are selective delignifiers can degrade polyphenols thanks to their chemical proximity to lignin. *Ganoderma* spp. are able to degrade polyphenolic compounds and show chemotropic growth towards these (Schwarze and Ferner 2003). The limited amounts of nitrogen in wood makes it resilient to decay (the C:N ratio of birch wood is typically of 55 and 401 for sycamore wood). Nitrogen is mostly found in parenchyma and is known to move towards the bark surface during log desiccation (Boddy 1992), which may make it particularly interesting for mycoboscus use. To fill in for a lack of available nitrogen, some white-rot fungi such as *P. ostreatus* have been shown to be able to predate nematodes, while the latter usually feed from their mycelia (Barron 2003).

The wide distribution of wood-decaying fungi suggests with increasing evidence that environmental factors and substrate specifics drive the outcome of fungal competition. Studies of lignicolous fungi ecological distribution point to water content and temperature variations being the principal drivers of selection (Boddy 1983)[25]. Furthermore, while the pH of wood is most often in the 4.0 – 5.5 range (beech: 5.11, birch: 5.29, for instance) (Geffert *et al.* 2019), it has been demonstrated that substrate pH is also influenced by the action of decay (Freschet *et al.* 2012). It has been reported by comparative transcriptonics that white-rot related genes of a *Phanerochaete carnosa* species would all express in fir (*Abies balsamea*), pine (*Pinus contorta*), spruce (*Picea glauca*), and maple (*Acer saccharum*) media, albeit to a different extent. For instance, MnP genes read was 2.42 times higher in fir media compared to maple, but LiP related genes read was 4.7 folds higher in maple (MacDonald *et al.* 2011). A variability in gene expression has also been reported for *P. chrysosporium* on aspen (*Populus grandidentata*), pine (*Pinus strobus*) and glucose-based media (Vanden Wymelenberg *et al.* 2011). Varying decay efficacy of *T. versicolor* on eight media ranging from quaking aspen (*Populus tremeloides*), basswood (*Tilia americana*), eastern white pine (*Pinus strobus*) and balsam fir (*Abies balsamea*) with mass losses ranging between 15 and 51 % has been reported for a twelve weeks decay period (Blanchette *et al.* 1988). This indicates both a resilience of fungal species to the use of a variety of available substrates, but also a precariousness considering the distribution of competitors of an individual species. This can lead to fungi chemically wrestling for resources (Hiscox *et al.* 2018). *Ganoderma* spp. have been reported to have the competitive advantage of being able to degrade reaction zones in trees (Schwarze and Ferner 2003; Shain 1967), although they are not the only ones (Schwarze and Baum 2000). In the wild, host tree species and their xylem vessel structure can have an influence on patterns of decay: the tree group that has the most diffuse vessel system can result in a slower initial colonisation. Gymnospermous wood having 3 – 4 mm long tracheids, benefits from a cellular compartmentalisation strategy to oppose fungal

[25] Climate change has then been related to shifts in fungal niches across hosts (Boddy *et al.* 2014).

Wood type	**Cell type**	**Layer**	**Share of total lignin content** (%)	**Lignin concentration** (%)
Gymnospermous early wood	Tracheid	S1 – S3	65	24
		ML, P	21	49
		Cell corners	14	64
Gymnospermous late wood	Tracheid	S1 – S3	75	22
		ML, P	14	51
		Cell corners	11	78
Angiospermous wood	Fibre	S1 – S2	60	19
	Fibre	ML, P	9	40
	Fibre	Cell corners	9	85
	Vessel	S1 – S3	9	25
	Vessel	ML, P	2	40
	Ray cells	S1 – S3	11	25

Table 2.6: Approximated lignin distribution in the layers of the wood cell wall. Adapted from Ferraz *et al.* 2014.

decay in addition to its guaiacyl lignin content. These topological effects are prominent at the early stage of fungal colonisation (Boddy 1992). Finally, in natural environments, fungal degradation happens in succession as the decay activity of primary, secondary and late colonisers compete and modify the composition of wood (Fukasawa and Matsukura 2021). This is called *substrate succession*, and might inspire multispecies-based mycoboscus production. The potential production of volatile organic compounds by fungi influences the attraction or repealing of insects in the wild (van den Brandhof and Wösten 2022) and decomposition succession. This could be investigated to support the design of mycobosci facilitating interspecies fostering.

2.6 Water in fungal decay

The presence of capillary water in the substrate is a prerequisite to allow extracellular transport of metabolites in fungi, and to establish osmotic potential for allowing turgor pressure and subsequent hyphal growth. Turgor also enables fungi to penetrate solid substrates (Money 1995). It has been reported that the white-rot species *Physisporinus vitreus* radial growth at pH 5 and 25 °C increased from just below 0.5 mm/d for a water activity $a_w \leq 0.974$, to 4.5 mm/d at $a_w = 0.998$ (Fuhr *et al.* 2011). The production of enzymes by fungi and their catalytic activity has been related to water activity, with varying local optima depending on the depressor that was used in the study, pointing to $a_w \geq 0.99$ for optimal catalytic activity here (Gervais and Molin 2003).
SSF requiring the absence of solution in between particles for a mycelium to develop, the substrate water holding capacity is an important parameter for ensuring an optimal fungal decay and growth (Brischke and Alfredsen 2020; Engelund *et al.* 2013; Gervais and Molin 2003). In timber, moisture can exist as bound water within cell walls below fibre saturation point (FSP), free liquid water in cell cavities above FSP, and water vapour. Relative humidity (RH) of up to 97 – 98 % is generally considered to correspond to cell wall uptake (hygroscopic range), and as RH moves toward saturation, wood cell void uptake tends to become more significant in the moisture distribution (over-hygroscopic range) (Thybring *et al.* 2018). The latter is considered as an optimal range for fungal growth in lignocellulosic SSF. The validity of this optimum is nonetheless discussed as it was shown that *T. versicolor* would colonise European beech wood at a minimum moisture content (MC)[26] of 15 % (equivalent to 75 – 80 % RH at –1 – 37 °C; R. J. Ross 2010 p.4–4) for a mass loss over 2 % (Meyer *et al.*

[26] Moisture content is expressed in this thesis as the ratio of water mass to dry substrate mass following the wood standard (R. J. Ross 2010).

2016), that brown-rot fungi grow at 92 – 97 % RH, and that instrumental limitations are encountered in the over-hygroscopic range thus complicating measures and experimental control (Thybring *et al.* 2018). The FSP of European beech wood has previously been determined to be 24.02 % MC at 97 % RH and 20 °C (**jankowska_determination_2016**). The minimum moisture threshold has been introduced as the metric having the most impact on predicting fungal growth (Brischke *et al.* 2017; Brischke and Alfredsen 2020; Thybring 2013). It represents the MC below which fungal decay cannot be initiated, and is typically given at a mass loss $\geq$ 2 %.
Hydroxyl groups are the predominant sorption sites for water molecules in wood (Engelund *et al.* 2013). The amount of available hydroxyls in hemicellulose is twice as high as in lignin, and four times higher than that of cellulose (Thybring *et al.* 2017). It has been reported that cellulose can endure structural changes while wood is desiccated (Atalla *et al.* 2009; Leppänen *et al.* 2011). In the literature, it has been hypothesised that lower desiccation rates would give enough time for amorphous cellulose to crystallise, rendering the substrate less hydrophilic. On the contrary, higher rates of desiccation may lead to more prominent amorphous cellulose and hydroxyl availability (Leppänen *et al.* 2011). Lignin, hemicellulose, and pectin interrupt cellulose crystallisation, which reflects on the higher crystalline content towards the S3 cell wall layer (Salmén 2015). Because hemicellulose has a softening point at room temperature and 18 – 20 % MC (Engelund *et al.* 2013), while lignin softens above 60 °C when saturated by water, and cellulose is hardly softened in its crystalline configuration ($\geq$ 200 °C), higher thermal treatments may result in hemicellulose degradation thus rendering the substrate less hydrophilic (Ramiah 1970; Werner *et al.* 2014).
Finally, the amount and quality of water in the SSF system varies during fermentation. Fungal oxidative decay of polysaccharides frees water

along with CO_2:

$$(C_6H_{10}O_5)_n + 6n(O_2) = 6n(CO_2) + 5n(H_2O) \tag{2.1}$$

$C_6H_{10}O_5$ being the repeating unit of glucose-based polymers, such as found in starch or cellulose. As bound water is also freed during decay, and while substrate dry mass loss is occurring, the MC is increasing if the system is closed. The activity and location of water in SSF systems are then necessary information to predict fungal development. They are of primary interest to design and produce mycobosci, yet, to the best of the author's knowledge, no study investigating the role of water in mycoboscus production has been reported to date. The optimal moisture content for laccase, LiP, and MnP activation is consistently reported to be 60 – 75 % (Hariharan and Nambisan 2013; Irshad and Asgher 2011; Iqbal *et al.* 2011; Asgher *et al.* 2010).

2.7 Regulating enzymatic activities

Mycoboscus protocols can be adjusted based on the chemical profile of locally available wood resources and selected fungal species to achieve various design goals. This can be approached by pH and temperature regulation, and monosaccharide, nitrogen, and metal ion supplementation. Optimal ligninase activity was reported for a pH of 4.0 – 5.0 at 30 – 35 °C (Hariharan and Nambisan 2013; Irshad and Asgher 2011). pH regulation can be achieved by addition of $CaCO_3$ in the substrate as a buffering agent to avoid acidification from CO_2 production. Gypsum supplementation has been reported in mycoboscus cultivation (Holt, Mcintyre *et al.* 2012). While its use has no impact on the substrate pH, it contributes to adding calcium to the medium. This is useful for pectate lyase activity if the wood has a low Ca^{2+} content, which acts on the 1 – 4 % share of pectin in wood. Varying the C:N ratio has also shown to lead to an optimum of 20:1 (Hariharan and Nambisan 2013). The form of the carbon and nitrogen rich additives has been reported to have an impact on specific enzyme activation: maltose and urea addition was reported to increase LiP and MnP production in a *T. versicolor* species, while glucose and yeast extract resulted in increased laccase production (Iqbal *et al.* 2011). A similar investigation with a *G. lucidum* strain reports that the use of beef extract increased LiP production by approximately 30 % as compared to using yeast extract, peptone, or urea, and sucrose had a significant effect on ligninase activity as per other sugar types (Hariharan and Nambisan 2013). Wheat bran is a frequently used substrate additive in SSF. It contains high contents of β-glucans, xylan, cellulose, lignin, B vitamins, and mostly: minerals (magnesium, phosphorus, manganese, zinc, and iron) (Hemery *et al.* 2007). The success of this supplement (Song *et al.* 2013) may be related to its mineral contents, especially manganese as it is a limiting compound for MnP and VP activity (Akpinar and Ozturk Urek 2012). Supplementation of substrates by $MnSO_4$ has been shown to result in an increase in ligninase activity in *S. commune* (Irshad and Asgher 2011)—most probably by synergistic action of enzymes. Biofuel production aims at efficient extraction of sugars from wood. Fungi that are selective delignifiers have been extensively investigated in this context. The addition of phenols has been reported to be considerably promoting ligninase activity (Mishra *et al.* 2017). Syringic acid has been shown to increase LiP and laccase activity, and gallic acid for MnP, as described in the same study. With an optimised substrate, the authors reported increases of 2.4 to 45.5 fold across the diverse ligninase activities, leading to a 2.3 factor improvement in lignin decay and 7.1 in selectivity value[27] (A. J. Chang *et al.* 2012). The addition of H_2O_2 to the substrate, necessary for ligninase activity,

[27] Selectivity is expressed as the ratio of lignin degradation over cellulose and hemicellulose degradation (A. J. Chang *et al.* 2012).

has also been investigated with significant results (M. B. Angelova *et al.* 2005; D. Li *et al.* 1995). $CuSO_4$ supplements have been shown to increase laccase activity (Mishra *et al.* 2017). The use of surfactants such as Tween-80 has been reported to increase ligninase permeation through the cell wall (Asther *et al.* 1987), enhance the solubilisation of lignin and improve hydrolitic activities (Sánchez Muñoz *et al.* 2021), and even proved useful more recently in promoting all ligninase activity, at a 4 wt% optimum for *G. lucidum* fermentation (Asgher *et al.* 2010). With an approximate 80 % content of starch, 12 % proteins, 3 % of other monosaccharides (arabinose, xylose, galactose, glucose), and traces of minerals (Andersson *et al.* 1993), white and whole wheat flour constitute useful additives to promote cellulase and hemicellulase activity. Moreover, the use of spent coffee ground is a widespread decay promoter in mycoboscus craft. It is composed of 39 % hemicellulose (predominantly mannose and galactose), 24 % lignin, 17 % proteins, 2 % fat, and 1.3 % ashes. Although it has a high lignin content, coffee grounds contribute to increasing nitrogen and mineral content in the substrate, in addition to glucose polymers. Coffee silverskin, a by-product of the roasting industry, has a quite different chemical profile: 24 % cellulose, 29 % lignin, 19 % proteins, 17 % hemicellulose (primarily xylose), and a remarkable 5 % ashes (Ballesteros *et al.* 2014). Phenolic compounds present in both substrates have been investigated for their anti-fungal abilities against white-rot fungi. Using a *T. versicolor* species, it was reported that the supplementation of 3 % coffee silverskin extract resulted in about 65 % growth inhibition, while 1 % caffeine led to a complete inhibition, and 1 % spent coffee ground in about 20 % inhibition (Barbero-López *et al.* 2020). This is related to the introduction of phenolic antioxidants such as caffeine, caffeoylquinic acid lactone, and feruoylquinic acid (Bresciani *et al.* 2014; Borrelli *et al.* 2004; Foti 2007). Furthermore, laccase treatment of lignocellulosic substrates has been shown to increase lignin antioxidant activity (Z. Li *et al.* 2018).

Finally, the addition of phosphorus and calcium in the form of superphosphate ($CaH_6O_8P_2^{2+}$) has proven to increase decay yield. Varying the supplementation from 0.025 wt% to a 0.3 wt% optimum in wheat straw, lignin degradation increased by a 4.5 fold, while cellulose, hemicellulose, and protein degradation increased by a factor of about 2.4 while enduring *P. ostreatus* decay (Tripathi and Yadav 1992). A higher optimum of 0.5 wt% superphosphate addition has been reported for alkaliphilic white-rot *Coprinus* spp. (Yadav 1987), and even 1.0 wt% with *T. versicolor* (Yadav and Tripathi 1991).

2.8 Mycelia mechanical properties

Mycelium acts as a binder between wood particles: it does so by degrading lignocellulosic compounds and producing a dense hyphal network that interlocks dispersed particles. The binding between two particles is not distinctly strong but the very high redundancy in the composite can result in a mechanically valuable material.

It has been reported that a *P. ostreatus* mycelium would be more than two times stiffer than a *G. lucidum* one when both cultivated on a cellulose medium (Haneef *et al.* 2017). Authors comment on this difference with regards to the higher protein and lipid content in *G. lucidum* that can act as plasticisers. Furthermore, in this referenced study, a set of experiments grown on a PDB-cellulose medium has been compared to the previously mentioned results, and it was found to decrease the Young's modulus of the *G. lucidum* mycelium film by a factor of 3 while increasing its elongation at break by 2.3-fold, although only lipids were higher in these samples as the protein content of *G. lucidum* cultivated on PDB-cellulose is lower than that of cellulose-cultivated specimens. This corroborates their previous comment on plasticisers, and suggests that nutrition medium tuning could help specify composites behaviour further than solely seeking stiffness. This referenced study suggests that the matrix phase of the composite can be made closer to an elastomer or stiffened by varying protein, lipids, or alcohol contents. Similar findings have been reported (Antinori *et al.* 2020). Isolated mycelium mechanical behaviour has been investigated so as to model it (Islam *et al.* 2018b). This referenced study relies on the characterisation in tension and compression of a pure mycelium. At the resolution of hyphae, the researchers have identified three main stages of response of a lignicolous mycelium: linear elastic behaviour at small strains (hyphae bending), fibre buckling and local structural collapse or densification at larger strains, and rapid stiffening associated with full compaction and large number of inter-fibre contacts. To fit their finite-element analysis (FEA) simulation model, a hyperelastic model was used in this study as mycelium undergoes large displacements in its elastic behaviour and results in a non-linear stress-strain response. Young's moduli situate in the 0.6 – 2.0 MPa range for a density in 30 – 50 kg/m^3, the yield stress in 40 – 80 kPa, and ultimate tensile strength in 100 – 300 kPa (Islam *et al.* 2017). It was reported that a *S. commune* species with suppressed SC3 hydrophobin gene resulted in a density increase between 1.4 and 3-fold to a wild strain, for a tensile stiffness increase between 2.3 and 4.4-fold in various light and CO_2 exposures (Appels *et al.* 2018). The stiffness of wild *S. commune* species was reportedly 15-fold higher than that of cellulose-cultivated *P. ostreatus*, while the former displayed a lower elongation at break. The reports are presented in the table 2.7.

2.9 Substrate mechanical properties

Most commercially interesting woods have a Young's modulus situated in the range 5.5 – 15.7 GPa at 12 % MC, and 4.4 – 12.3 GPa when freshly cut (green) (D. W. Green *et al.* 1999). The distribution of these performances is similar in gymnospermous and angiospermous woods. Wood

Tensile behaviour	$SC_{\Delta sc3}$	SC	PO*	PO**	GL*	GL**	ND†
Young's modulus (MPa)	1237 – 2727	438 – 913	28	17	12	4	0.6 – 2.0
Ultimate strength (MPa)	15.6 – 40.4	5.1 – 9.6	0.7	1.1	1.1	0.8	0.1 – 0.3
Elongation at brake (%)	1.7 – 2.6	1.2 – 1.4	4	9	14	33	–

Table 2.7: Tensile behaviour of mycelia from *S. commune* ($SC_{\Delta sc3}$, SC) (Appels *et al.* 2018), *P. ostreatus* (PO) (Haneef *et al.* 2017), *G. lucidum* (GL) (Haneef *et al.* 2017), and a non-disclosed species (ND) (Islam *et al.* 2017). *Cellulose medium. **Cellulose and PDB medium. †Non-disclosed medium.

particles are therefore important load-carrying members of the composite system as compared to the mycelia elasticity discussed previously, and reduce the magnitude of stress experienced by the mycelial matrix. The plastic strain of the composite is contributed to only by particles in such a composite. The angular shapes of wood debris contribute to crack initiation (beech wood aggregates used in experimental studies are presented in Fig.3.45) and the dewetting behaviour of the larger particles present in a mycoboscus composite is a principal contributor to damage nucleation in two-phase particulate composite, although they contribute to composite stiffening by matrix shielding (Lu *et al.* 2020). Furthermore, the shape, nature, and distribution of particles in two-phase particulate composites has been shown to have a substantial influence over the load transfer between members and hence their overall stiffness (Shen *et al.* 1994). While lignin is a primary contributor of strength parallel to grain, hemicellulose supports compressive strength perpendicular to grain. Its decay greatly affects the structural integrity of wood and its hardness (Curling *et al.* 2002).

In particle studies of granular materials, shape parameters such as flakiness/flatness (ratio of particle thickness to width), elongation (length to width ratio), sphericity (deviation from a sphere geometry), and roundness/angularity (measure of angles sharpness) have been investigated (Fig.2.14; Selig and Roner 1987). Without matrix phase[28], studies have reported that a 3:1 ratio of flaky to bulky particles content would be an approximate optimal for shear strength (depending on the system of study). This was related to cohesion increase under stress related to particles interlocking. Furthermore, it was reported that a higher particle angularity leads to a decrease in elastic modulus, while ultimate strength increases. The shear strength was reported to follow the same dynamic, increasing with particle angularity. Increasing flakiness and angularity leads to increased cohesion. It is also subjected to a higher abrasion and leads to damage accumulation under repeated loads, thus leading to strain accumulation (Selig and Roner 1987).

While wood particles are of common use in mycobosci, other more elastic substrates have been used such as non woven cotton fibres (Appels *et al.* 2019). A few researches have also been investigating the addition of non-organic aggregates to a lignocellulosic substrate for improving its stiffness, such as carbonate sand (Gou *et al.* 2021) (which also acts as a pH buffer being mainly composed of $CaCO_3$), and sand and gravel (Moser *et al.* 2017). It was reported that supplementation of a cotton and wheat bran medium with 37.5 wt% carbonate sand resulted in a 1.6 fold increase in stiffness, and a factor 4 increase in ultimate compressive strength, while density increased by 27 %.

[28] The stiffest reported pure mycelium of a *P. ostreatus* strain in section 2.9 had a Young's modulus of 28 MPa, 178 – 534 times more elastic than woods (D. W. Green *et al.* 1999).

2.10 Mycobosci mechanical properties

The large number of influential variables on the mechanical properties leads to the emergence of a variety of methods for tuning them. While common practice leads to a median compressive modulus of 3.91 MPa (see table 2.8 and Fig.2.15), post-cultivation heat-pressing has been reported to lead to Young's modulus across the range of 35 – 97 MPa and flexural modulus across the range of 34 – 80 MPa (Appels *et al.* 2019). The use of the particle-additives strategy, additionally to a wheat bran supplemented medium made of cotton stalk, has proven to have a significant effect: an elastic modulus of 48.5 MPa was reported for a 37.5 % carbonate sand addition in *P. ostreatus*-inoculated specimens, while a non supplemented control in this study reached 30.3 MPa (Gou *et al.* 2021). A second study reported on an elastic modulus situated in the 39 – 60 MPa range for a density in 240 – 265 kg/m^3 (Z. Yang *et al.* 2017). Authors of the latter study are elusive about production details, but nonetheless they indicate the use of Alaska birch shavings (*Betula neoalaskana*), millet grain, wheat bran, calcium sulphate, and the addition of a natural fibre. The specifics of the performative batch are not disclosed, but a precolonisation was performed prior to moulding. The performance increase does not come from a single factor considering the experimental plan. The considerable use of fibres (of undisclosed quality), grains, brans, and $CaSO_4$, leads to understanding this as an hybrid improvement using both substrate based reinforcement with the fibres, and additives. Other strategies to effect increases in stiffness have been investigated, such as with the introduction of SBR Latex, a bonding agent for the construction industry. With a 5 % addition of SBR Latex and 0.5 % silane coupling agent to a cotton seed hull medium, the cultivation of a *P. ostreatus* strain displayed a 2.39 factor increase in yield strength with a 27.6 % density increase (He *et al.* 2014). The environmental impact of the design was not discussed in the study. The addition of another material with high environmental impact (Arvidsson *et al.* 2015), cellulose nanofribrils (CNF), has been reported (W. Sun *et al.* 2019). Between two specimen groups, the optimal supplementation of CNF was of 2.5 – 5 wt% with a contribution in Young's modulus of a factor 2.56 – 5.6 and 2.8 – 7 in ultimate strength. In this reported study, the effect of densification on composites embedding a 2.5 wt% CNF part was reported: densifying from 300 kg/m^3 to 600 kg/m^3 contributed to increase the yield strength exponentially by a factor 17, while the stiffness increased 14.38 fold.

The elastic modulus in tension of mycobosci is reportedly situated in 3.0 – 13.0 MPa. These values result from a densified *T. versicolor* mycelium, cultivated for 24 days on wheat bran supplemented beech sawdust (Appels *et al.* 2019). For this specimen group, a flexural modulus of 9.0 MPa was reported. The state-of-the-art in mycoboscus research largely considers monolithic and homogeneous composites, with the exception of a few study groups investigating jute type materials in sandwich composite reinforcement and composition with wood panels (L. Jiang *et al.* 2017; Sàez *et al.* 2021; Ziegler *et al.* 2016).

Across the state-of-the-art, three main design strategies can be identified: *densification* (by dense packing, cold or hot-pressing), *composition* (by introducing macro structuring elements such as fibres, and aggregates selecting), and *supplementation* (by chemical tuning of the substrate). The contribution of these strategies on elasticity and strength are illustrated in Fig.2.16. These strategies are substrate-focused but target the tuning of mycelium properties in the case of supplementation, while the very choice of the fungal species can result in both different cultivation lead time and mechanical properties. Genetic modification of fungi could promote specific enzymatic activity or modify the composition of its cell wall to help improve performances, while the investment cost and ethics of this practice may lead to prioritising more energy-efficient and affordable techniques.

2.11 Mycoboscus Life-Cycle Assessment

Three mycoboscus Life-Cycle Assessments (LCA) have been conducted recently. Looking to the manufacturing of a standardised brick design (DIN EN 771-1) from hemp shives colonised by a *F. fomentarius* strain, and investigating a cultivation from the making of a petri dish culture to the brick via rye grain cultivation, an account of global warming potential, acidification potential, eutrophication potential, photochemical ozone creation potential, soil quality potential and user deprivation potential was done (Stelzer *et al.* 2021). The authors report that the electrical use for sterilisation of substrates and drying of the cultivated bricks were the most significantly impacting categories along with rye grain and hemp acquisition. The German consumption electricity mix was used in the study. Because of the higher share of coal use in Germany, 81.4 % of the CO_2 eq. of the global warming potential for producing a brick was contributed to by electricity use. This represented 58.7 % of the acidification potential and 73.2 % of the photochemical ozone creation potential. Hemp use weighted to 95.2 % of the eutrophication potential of manufacturing along with 84 % in water scarcity. Changing substrate from hemp to rapeseed straw resulted in a 33.4 % increase in acidification and 21.1 % eutrophication potentials, which is related to different fertiliser use in the cultivation of rapeseed. Land use estimate sees a decrease of 17.3 % in this scenario. Poplar wood use, compared to hemp, would result in a 49.4 % reduction in eutrophication as no fertilisers are needed, but would increase land use by 31.4 %. At an industrial scale scenario, the production of a hemp shives-based brick as compared to a concrete one would see a potential halving of acidification potential, climate change potential and water scarcity potential, and a reduction by almost a factor 5 in smog po-

	Compressive strength (MPa)	**Tensile strength** (MPa)	**Flexural strength (MOR)** (MPa)	**Compressive Young's modulus** (MPa)	**Tensile Young's modulus** (MPa)	**Flexural Young's modulus** (MPa)	**Density** (kg/m^3)
Count	98	17	110	68	16	116	183
Mean	0.55	0.25	6.04	11.84	120.91	512.36	290.61
S.d.	0.94	0.46	4.76	17.54	298.75	398.42	182.00
Min	0.03	0.01	0.05	0.14	0.52	0.40	23.00
25 %	0.17	0.04	0.94	1.19	3.24	49.60	168.85
50 %	0.28	0.10	5.57	3.91	6.64	575.00	250.00
75 %	0.64	0.15	10.78	12.50	41.00	853.25	357.00
Max	7.99	1.72	15.30	71.77	1100.00	1460.00	1208.38

Table 2.8: Statistical distribution of selected mechanical characteristics for mycobosci.

tential. Such a hemp brick would see a 300 % increase in eutrophication potential due to fertiliser use, and 3800 % increase in land use compared to a concrete counterpart due to organic material use.
A higher contribution to global warming potential by the drying phase, 58.7 % of the total energy use in the process, was reported (Carcassi *et al.* 2022). The first study reported that drying represented 49.8 % of the electrical consumption of the cultivation process (Stelzer *et al.* 2021). Another study found that the incubation phase represented 73 % of the total electrical consumption and the drying phase 8.1 % (Livne *et al.* 2022). With a focus on accounting for biogenic CO_2 resulting from photosynthesis of the principal substrate and the metabolic CO_2 released by fermentation, the authors point to the possibility for a mycoboscus, as cultivated in their study, to be CO_2 eq. negative. This potential is highly sensitive to the incubation time, as the authors point to the fact that above two weeks of incubation, the composites should result in an emission of 45.7 kg CO_2 eq. m^{-3}. The substrates that were used in this reported study were recycled cellulose fibres and rapeseed straw, the latter necessitating fertiliser use during its cultivation and therefore impacting negatively other environmental metrics. These raw materials were considered as waste and therefore represent a negative carbon emissions equivalence in the calculation. With a similar manufacturing process, the study using bamboo found that only with a fully renewable sources-powered electricity mix would a mycoboscus, as defined in the study, be environmentally competitive to kenaf, cork, cotton stalk or straw solutions for insulation (Carcassi *et al.* 2022).

Although the studies reported variable electrical consumption estimations, the three clearly point to the high sensitivity of mycoboscus manufacturing to electrical consumption and thus to local energetic mixes which may imply the use of fossil fuels. Future developments in manufacturing methods could look to reduce or nullify energy consumption at the stage of sterilisation, incubation and drying. While a study points to this need to reduce the energy use in drying (Carcassi *et al.* 2022), the necessary inactivation of chlamydospores (under environmental conditions that have yet to be investigated; see section 2.2.1) will require a future increase of energy use nonetheless. The provenance of the principal substrate is also shown to be of a high importance in the LCA as fertilisers may be in use, and the agricultural nature of the materials implies a higher land use potential than that of ceramic materials—although mycobosci perform better on all investigated metrics and at scale, with the exception of eutrophication potential. The practice of upcycling byproducts from other supply chains may help to extend the life cycle of materials that would otherwise be discarded, while the LCA of the processing of these byproducts to render them workable in mycoboscus manufacturing will be useful to drive design decisions.

2.12 Conclusion

Three design strategies were identified from the literature review: supplementation, densification and composition. From the enzymes review it can be understood that resulting substrate fitness and mycelia mechanical behaviour can be adjusted by means of supplementation. This can be approached by pH and temperature regulation, and sugars, nitrogen and metal ions supplementation. The properties of mycelia have furthermore been shown to be influenced by such. Use of compounds such as pH buffer $CaCO_3$ could be systematically investigated in the future. Lignocellulosic substrates selection can also have a significant effect, unsupplemented softwoods being theoretically chemically unfit for optimal white-rot decay. The *D. squalens* species may be investigated in the future for its preference for white-rot decay of softwoods. Use of sycamore or ash wood could be investigated in the future as they display higher syringyl monomer contents and would theoretically be fitter for white-rot decay with species showing a substrate preference for hardwoods. The C:N content of sycamore being 401, supplementation may be necessary for optimal fermentation. The effect of water activity upon mycoboscus colonisation-related performances has not been reported yet in the state-of-the-art. Considering reports in SSF studies of several fold increases in colonisation rates, investigating its effect in mycoboscus may result in improved lead production times and performances. Moreover, qualifying the effect of hydroxyl group availability in hemicellulose and cellulose depending on previous wood processing could lead to further medium optimisation. This particular aspect is expected to be specifically useful for paste-like substrate design, such as found in mycelium-related 3D printing.

Stirring the focus of future research towards such specific aspects may result in a higher barrier to entry of a technique that has a strong vernacular interest. The use of Fourier-Transform Infrared (FTIR) spectrometry (section 3.2.3) by experts within a network of citizen practitioners could be useful to adapt cultivation strategies in a constructive manner: the qualification of an available substrate may lead to specifying necessary processing for it, supplementation, species selection and relevant composition strategies. While industrial and academic research may result in defining a panel of protocols for a selection of performative materials, constructive practices informed by locally available materials could coexist.

As the species in use are versatile through their capacity of expressing a wide array of enzymes, the mycoboscus craft is affordable and forgiving should the cultivation conditions be sub-optimal. LCA studies point to the higher impact on eutrophication, acidification and land use (soil quality potential) of mycoboscus systems compared to mineral solutions; the enzymatic versatility allows for preventing the tying of emerging mycoboscus practices to agricultural supply chains for other than wastes.

The upcycling of a range of cellulose-based wastes such as discarded textiles or other organic byproducts is a possible, mechanically promising and environmentally recommendable development path. To provide cultivation conditions for the fungus of use, chemicals and substrate blends will need to be elaborated from these sourcing opportunities.

59 species have been inventoried across the referenced mycoboscus literature, of which 15 are considered in the state-of-the-art of the present study, focusing on mechanical characteristics (table 2.1). Just as mycoboscus commercial solutions are advertised for their compostability (Ecovative 2022) and biodegradation studies are conducted (Van Wylick *et al.* 2022), the large scale diffusion of potential chlamydospores in ecosystems could cause ecological imbalance due to the life-history strategies of the considered species as they can be opportunistic pathogens or potential parasites (tables 2.1, 2.2 and 2.3), and because they generally benefit from the ability to synthesise potent enzymes, this makes them ecologically competitive. Along with the prevention of fruiting body formation in the mycoboscus cultivation process for health risk prevention for the craftsperson, the inactivation conditions of chlamydospores of species in use need investigating before the products see an increase in manufacturing volumes and are widely adopted. Furthermore, the selection of less performative species (as per manufacturing efficiency) to try and prevent a possible ecological imbalance in the case of a lack of control of the lifecycle seems to be a relevant development effort. This is a conclusion supported by experiments conducted in laboratory conditions having shown a significant effect of primary decomposer assemblies on subsequent decomposition activity and population over a five month period (Fukasawa and Matsukura 2021). Primary decomposer distribution and decay type has also been shown to influence seedlings density (Fukasawa 2012). The conclusions of these studies invites the scoping of indigenous species for mycoboscus within localised supply chains instead of using globally

standardised highly performing or genetically selected or modified species, out of a principle of ecological caution.
More species than the 59 listed can be good candidates for future mycobosci recipes. *A. delicata*, for instance, would be an edible species to evaluate experimentally considering its wood rot capability. This species is supposed to have the capacity to form chlamydospores (Buchalo *et al.* 2011). In general, local species for local uses are ecologically recommended (*local-to-local*), which may support a non-scalable crafts manufacturing network establishing. As introduced, this may reflect the acknowledgement of persisting frailties in the technical system regarding a local supply potential variability but may be compensated by other stiffening strategies or the development of organisational modes of compensation, and will reflect ecological traits influenced by geography and climate.
The biochemical profiles found in plant-based substrates used as the dispersed phase for the composite material are both influenced by climate and seasons (Zhang, Wang *et al.* 2021; Wang *et al.* 2016), and soil profiles (Passioura 2002; Russell 1912) among other factors. Lignin presence and geometrical configuration of wood cell wall are defining for the fungal growth rate to the fungal species. White rot species will better degrade angiospermous trees as their guaiacyl lignin content is lower than that of gymnosperms. Many published studies in the field of mycobosci do not inform on the accurate nature of their substrate; for mechanical studies, the size, shape and nature of those is also necessary to allow reproducibility of results. Furthermore, mycelium mechanical properties such as tensile elasticity have been shown to be influenced by protein and lipid content in the culture medium.

The LCA accounts point to the relatively large electrical consumption required to produce a mycoboscus employing current protocols—implying sterility and controlled incubating and drying. This aspect makes the material system non-competitive environmentally for insulation as compared to straw for instance (Carcassi *et al.* 2022). The replacing or excluding of needs for electrical appliances can make mycobosci competitive, for instance by using solar ovens to dry and inactivate them effectively. Developing working methodologies towards a low-energy or passive framework may support another cap on scalability again, which may promote the consideration of seasonality additionally to geographical situating. Non-sterile cultivation methods can be used with little protocol adaptation, for instance by increasing the ratio of inoculant to virgin substrate. The effect of situated fermentation practices on miso microbial colonies has been demonstrated previously by metagenomics (Evans 2022). This is a promising technical and cultural research track for mycobosci considering the varied contexts of production between workshops and high-end laboratories. A first metagenomic study was conducted in the context of mycoboscus production with non-sterile virgin substrate use (McBee *et al.* 2021).

Principles for supplementing or designing the chemical profile of the substrate have been presented, and the future opportunistic use of byproducts as substrate will necessitate the democratisation of affordable analytical methods to qualify local sources chemically and mechanically to obtain relevant recipes. The analysis of the byproducts will need to focus on describing cellulose contents or saccharides for carbon source, nitrate, ammonium or protein content for nitrogen, phosphate and phosphorus forms, sulphur content, and metal ions. An account of most of these compounds in a selection of byproducts is presented in table 2.9.

The densification strategy necessitates hot or cold pressing and intense electrical energy consumption. Densifying with a mechanical press that is human-powered or gravimetric seems relevant to avoid making mycobosci an environmentally costly technical system, although cold pressing in this manner may involve lower final strength and stiffness of the products. The high variety of substrates shapes that can be used, from dust to textiles and other forms, and considering the diversity of aggregates, represents an opportunity to passively modify the mechanical behaviour and efficiency of mycobosci. The composition strategy is scarcely evaluated in the state-of-the-art, both per macro structuring and aggregate specifics, although it presents a viable method to limit the embedded energy in mycobosci all the while integrating other forms of materials found opportunistically. Composition of aggregates can serve as a way to modify and control the composite behaviour under stress, and its macro structuring can serve to integrate reinforcements and functions. In the following chapter, experimental studies focusing on composition strategies, and their effect on compression, tension and flexion, are presented.

A visual synthesis of the mycobosci model of biochemical and mechanical parameters is presented in Fig.2.17 and as annotated in appendix A.1. As per supplementation, surfactant use, C:N ratio, Mn^{2+}, Ca^{2+} and $CaCO_3$ use, and temperature ranges are identified as the most significant variables to be qualified against a species and substrate opportunity. The inner crown thus represents wood cell wall and the principle effects of these variables on its composition. Moisture content is also identified as critical, and the outer crown indicates some common design parameters for composition. At fungi cell scale, the membrane, enzyme and proton pump expression are identified as important pathways that are influenced by the substrate profile and hence mycoboscus result.

Properties	Oak sawdust	Hornbeam sawdust	Tea waste	Wheat bran
OC (%)	56.79	57.78	51.96	55.23
C:N	162.26	525.27	23.09	22.09
Total N (%)	0.35	0.11	2.25	2.50
P (ppm)	149.08	0.00	2000.00	7650.77
K (ppm)	728.78	546.08	5110.00	4983.33
Ca (ppm)	2186.60	1179.52	9590.00	73268.00
Mg (ppm)	223.72	178.81	400.00	2180.93
Fe (ppm)	174.37	49.00	999.00	49.03
Mn (ppm)	70.35	37.99	1255.50	81.20
Zn (ppm)	1.93	3.61	301.40	48.83

Table 2.9: Mineral contents, organic carbon (OC) and C:N ratio of some waste materials. Adapted from Peksen *et al.* 2011.

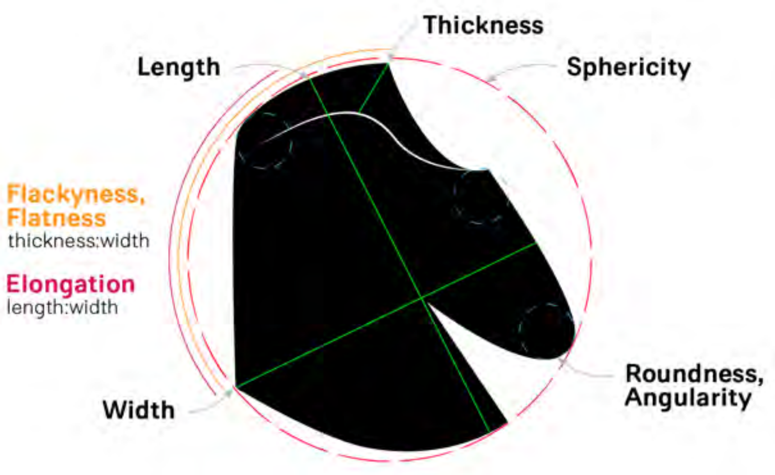

Figure 2.14: Illustration of common shape parameters to qualify aggregates.

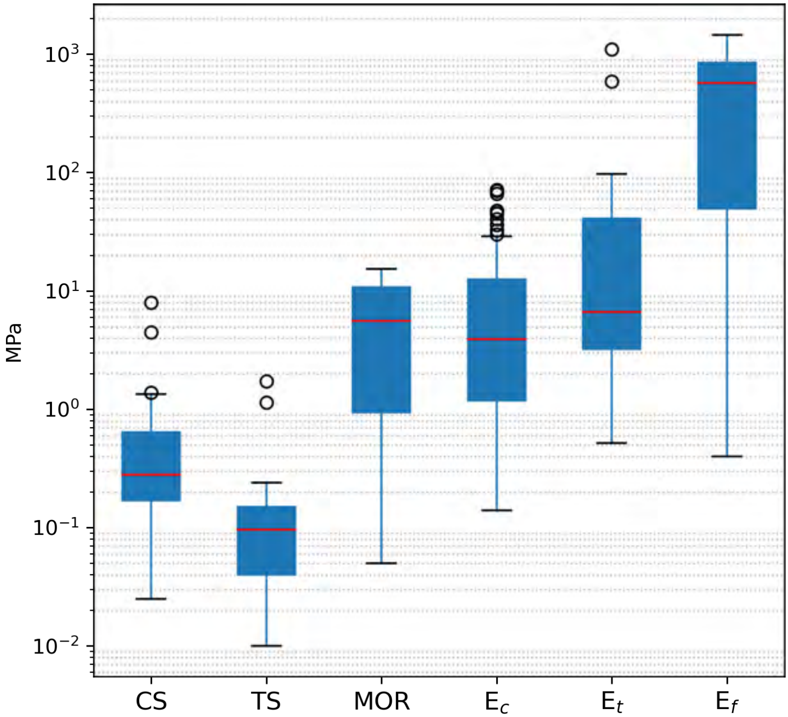

Figure 2.15: Boxplot of selected mechanical characteristics for mycobosci. CS: compressive strength, TS: tensile strength, MOR: modulus of rupture (flexion), E_c: compressive elastic modulus, E_t: tensile elastic modulus, E_f: flexural elastic modulus.

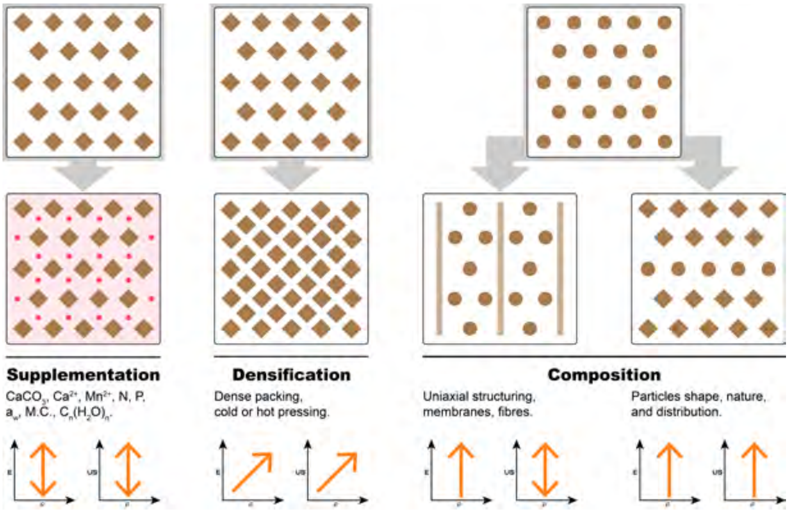

Figure 2.16: Effect of design strategies on mycoboscus elasticity and strength properties.

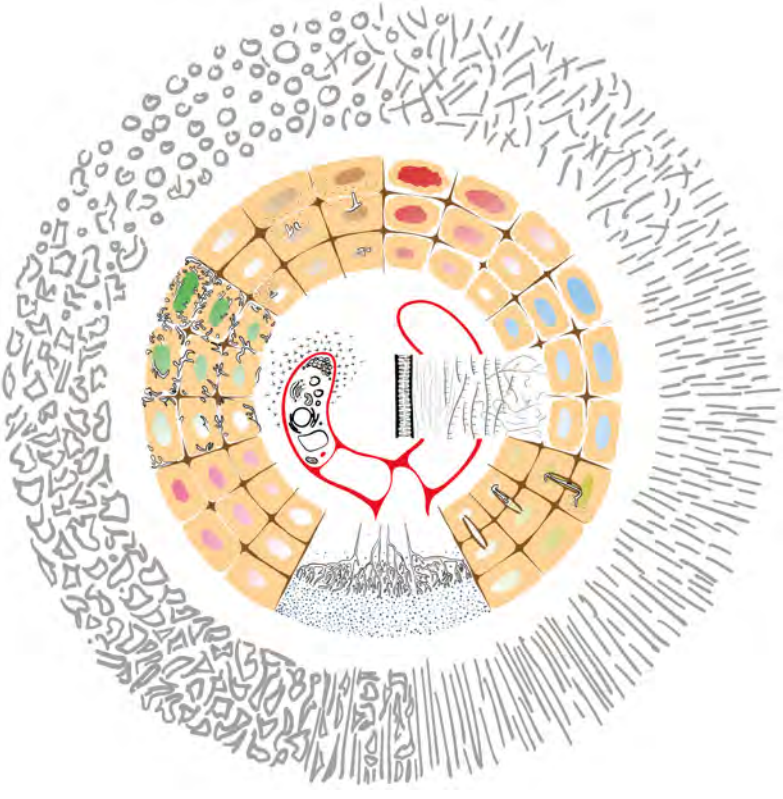

Figure 2.17: Multi-scalar mycoboscus model (2022). Annotated version: appendix A.1.

Chapter III – Composition Strategies

3.1 Introduction

Because mycoboscus cultivation protocols can be based on virtually any substrate containing organic polymers such as hemicellulose and cellulose or mono and disaccharides, and as a range of widely available agaricomycetes can be utilised, this class of composite shows potential in obtaining viable products for a variety of uses while resorting to opportunistic substrates in the form of byproducts. As two-phase particulate composites, isotropic mycobosci are made of a matrix phase that corresponds to a continuous and foam-like or elastomer-like mycelium and a dispersed phase composed of particles or fibres. Comparing the reported elasticity of mycelia in section 2.8 with wood ones from section 2.9, it can be noticed that the species in common use, such as *P. ostreatus* and *G. lucidum* have a tensile Young's modulus in the 4 – 28 MPa range, while woods have a Young's modulus in the range of 5.5 – 15.7 GPa. The aggregate phase is therefore much stiffer than the matrix—although fungal decay of lignin and hemicellulose softens the aggregates. The inner structure of a typical mycoboscus is presented in Fig.3.18.

A first understanding of significant variables for mycoboscus design is presented in section 2.9, where particle parameters such as flaki-

Figure 3.18: Typical section of a mycoboscus (2021).

ness/flatness, elongation, sphericity and roundness/angularity were introduced as important qualities. As represented with the densification strategy in Fig.2.16, the concentration of aggregates is an influential parameter on the composite mechanical properties (Appels *et al.* 2019). The arrangement and orientation of aggregates have an influence, which opens to considering two principal classes of aggregates: particles and fibres. The composite topologies that are considered in this work are presented in Fig.3.19. Structural composites as combinations of composite and homogeneous materials are positioned in the figure for reference, but were not studied. Dispersion-strengthened particulate composites concern aggregates of a diameter situated in 0.01 – 0.1 μm (Callister 1999 p.523). The interactions between the matrix and dispersed phases are happening at the atomic or molecular level for this class. The scale of aggregates used in the work presented here, and across the state-of-the-art, is $\geq$ 0.5 mm and includes negligible fractions of wood dust (10 – 30 μm). The composites that were focused on can thus be classified as large particle reinforced. Short fibre materials such as cotton stalks, corn stover or hemp shives were not studied.

Building a study focus on large particle composites and the methods to significantly modify their characteristics by way of composition, the chemical fitness of the principal substrate to the selected *G. lucidum* strain is evaluated in the first place. As this is confirmed experimentally, the effect of particle size for the retained European beech wood substrate is qualified under compressive and tensile stress response. Having determined the optimal aggregate size, macro composition strategies are evaluated as a means to modify the elastic and failure behaviour of mycobosci. This study is subsequently continued for flexural stress. A last experimental study is presented, with a focus on evaluating the effect of species selection and lignin type on the composite response under compressive load. Finally, the *G. lucidum* mycelium synthesis and binding capacity is

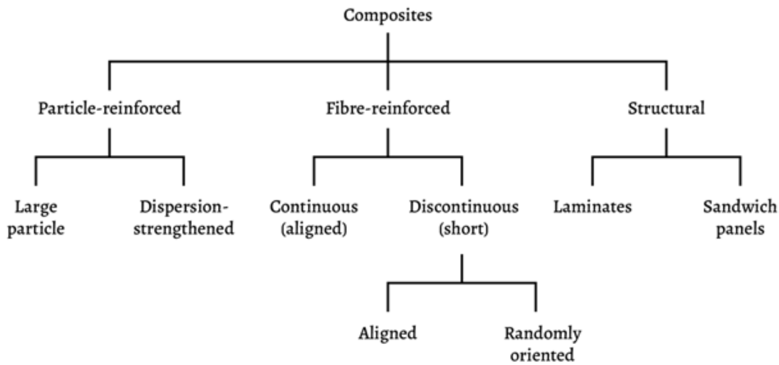

Figure 3.19: Typology of composites considered in this study. Adapted from Callister 1999 p.522.

determined analytically based on the rule of mixture for composites and experimental data[29].

The experimental works reported in this study thus concern:

- Decay chemical characterisation,
- Effect of particle size (compression & tension),
- Effect of composition (compression & flexion),
- Effect of fungal species and lignin in compression,
- Effect of particle size on aerobic respiration.

[29]The chemical study and compressive behaviour characterisation looking into aggregate size and composition strategies was the topic of a research disseminated in Rigobello and Ayres 2022; appendix D.1. The flexural behaviour characterisation was reported in Rigobello, Colmo *et al.* 2022 but without the data generated from the testing of specimens manufactured by Mogu; appendix D.2.

3.2 Materials and methods

Because *G. lucidum* is considered a very versatile ligninolytic fungus, in that it can exploit various strategies for the breakdown of lignin and can ultimately degrade all components of lignocellulose (section 2.3), it was selected for implementing the experimental plan. European beech wood and plant-based materials used to manufacture composites have been chemically qualified by Fourier-Transform Infrared (FTIR) spectrometry. In the same section, the decay activity of *G. lucidum* on beech wood is quantified by FTIR. European beech wood was selected as a principal substrate across experiments for its theoretical fitness to white-rot decay (section 2.9). Because the effect of substrate particles size has not yet been investigated experimentally for mycobosci, its effect on composite compressive and tensile stress response was evaluated. The effect of composition strategies over the mechanical behaviour in compression and flexion were then investigated with the best performing aggregate size. With a standard protocol, a change in strains was studied on the best performing granulate size between a *G. lucidum* and a *G. resinaceum* species. Similarly, and as a way to verify the impact of the nature of lignins on white-rot fungi colonisation experimentally, specimens of pine wood cultivated *G. lucidum* were tested under compressive stress. Based on the experimental results of isotropic controls from flexural, compressive and tensile studies, and by way of the rule of mixture for determining a two-phase composite Young's modulus, the specific matrix modulus was then approached.

A variety of experimental designs are being used in the field of mycobosci research to evaluate mechanical characteristics. Few studies consider material evaluation standards, and systematically derive from the guidelines of these standards (regarding specimen or experimental design). Among them, ASTM D3501 for wood-based structural panels in compression has been referenced (Elsacker *et al.* 2019), a standard designed for plywood, wafer-board, orientated strand board, and composites of veneer and of wood-based layers, with use of 2:1 (D:h) cylindrical specimens in the study, instead of rectangular cross-section as the standard advises; ASTM D695 for rigid plastics was also referenced (Sivaprasad *et al.* 2021), with a recommended 1:2 (D:h) ratio for cylindrical samples, but used with a diameter of 100mm and thickness of 23 mm in the study; ASTM C67 destined to brick and structural clay tile was referenced in a comparative study against clay bricks (Ongpeng *et al.* 2020), but without following the standard recommendations; and ASTM D2166-13 for cohesive soil was referenced (Z. Yang *et al.* 2017), but deviating from the standard in the study. In requiring the largest particle to be smaller than one tenth of the specimen diameter, the latter ASTM exemplifies the instrumental role these standards can play in systematically investigating materials based on their composition. The variety of specimen geometries found in the state-of-the-art and lack of consistent recourse to the standards challenges the portability of, and comparability between, experimental results. The referenced standards in the state-of-the-art are presented in table 3.10. The ASTM that is the fittest for evaluating these properties with adopting the two-phase particulate composite model is ASTM D1037 (ASTM 2020). It has the benefit of being the most referred set of guidelines in mycobosci development and covers various tests.

3.2.1 Cultivation method

Materials

Millet-grown spawn of species *G. lucidum* (reference M9726) and *G. resinaceum* (reference M9732) were acquired from Mycelia BVBA (Nevele, Belgium). The spawns were stored at a constant 4 °C and 65 % RH. The prin-

Test	Standard	Designation	Ref.
Compression	ASTM D3501	Standard Test Methods for Wood-Based Structural Panels in Compression.	Elsacker *et al.* 2019
	ASTM D695	Standard Test Method for Compressive Properties of Rigid Plastics.	Sivaprasad *et al.* 2021
	ASTM C67	Standard Test Methods for Sampling and Testing Brick and Structural Clay Tile.	Ongpeng *et al.* 2020
	ASTM D2166	Standard Test Method for Unconfined Compressive Strength of Cohesive Soil.	Z. Yang *et al.* 2017; Houette *et al.* 2022
	ASTM D1037	Standard Test Methods for Evaluating Properties of Wood-Base Fiber and Particle Panel Materials.	Ziegler *et al.* 2016; Chan *et al.* 2021
	ASTM C165	Standard Test Method for Measuring Compressive Properties for Thermal Insulations.	Holt, Mcintyre *et al.* 2012
	EN 826	Thermal insulating products for building applications – Determination of compression behavior	Elsacker *et al.* 2021
	ASTM D3574	Standard Test Methods for Flexible Cellular Materials—Slab, Bonded, and Molded Urethane Foams	Heisel *et al.* 2018
Flexion	ASTM C203	Standard Test Methods for Breaking Load and Flexural Properties of Block-Type Thermal Insulation.	Holt, Mcintyre *et al.* 2012
	ASTM C393	Standard Test Method for Core Shear Properties of Sandwich Constructions by Beam Flexure	L. Jiang *et al.* 2017
	ASTM D7250	Standard Practice for Determining Sandwich Beam Flexural and Shear Stiffness	L. Jiang *et al.* 2017
	ASTM D1037	Standard Test Methods for Evaluating Properties of Wood-Base Fiber and Particle Panel Materials.	Lokko *et al.* 2016; Sun and Tang 2012; Chan *et al.* 2021; W. Sun *et al.* 2022
	ISO 16978	Wood-based panels – Determination of modulus of elasticity in bending and of bending strength.	Elsacker *et al.* 2021
	ISO 12344	Thermal insulating products for building applications – Determination of bending behavior.	Elsacker *et al.* 2021
Tension	ASTM D1037	Standard Test Methods for Evaluating Properties of Wood-Base Fiber and Particle Panel Materials.	Ziegler *et al.* 2016; Chan *et al.* 2021

Table 3.10: Referenced standards for mechanical property determination in the mycobosci state-of-the-art.

cipal substrate of the specimens is European beech wood (*Fagus sylvatica*). Three granulations were used (BS: small, BM: medium, BL: large): 0.5 – 1.0 mm (Räuchergold type HB 500/1000, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany), 0.75 – 3.0 mm (Räuchergold type HB 750/2000, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany) and 4.0 – 12.0 mm (Räuchergold type KL 2/16, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany). A fourth particle type was added to the experimental plan, as a 1:1:1 volume ratio mix of the three granulations (BSML). Longitudinal fibres were introduced in a specimen series by using common reed fibres (*Phragmites australis*; Tækkemand Chresten Finn Guld, Køge, Denmark). 6 mm diameter rattan fibres have been used for compression and flexion testing (*Calamus manan*; B.V. INAPO, Bloemendaal, Netherland), such as hemp-based hessian (*Cannabis sativa* subsp. *sativa*; NEMO Hemp jam web 370 g/m^2, Naturellement Chanvre, Echandelys, France). The materials are illustrated in Fig.3.20.

Protocol

The MC within substrate cell walls is critical for understanding and predicting fungal activity (section 2.6). In mycoboscus production, lignocellulosic substrates composed of particle or fibres usually have a MC that is homogeneously prepared at 55 – 70 % (Appels *et al.* 2019; Elsacker *et al.* 2019), and the use of mineralized to sterile demineralized water has been documented as a means of moisturising (M. Jones *et al.* 2020; Appels *et al.* 2019; Elsacker *et al.* 2019).

The principal substrates, fibres and hessian were prepared at 70 % MC with mineralized water and sterilised at 121 °C for 15 min. The principal substrates were mixed with 16 wt% spawn and incubated in polypropylene filtered bags (SacO2, Deinze, Belgium) for 7 days at 25 °C in the dark. Once colonised, the principal substrates were broken down and formed with the sterile fibre and hessian into alcohol cleaned aerated PETG moulds.

Figure 3.20: Materials of the experimental studies (left to right, top to bottom): beech large (BL), beech medium (BM), beech small (BS), birch shavings (not used), hemp shives (not used), rattan fibres, hessian, and common reed fibres (2021).

The formed specimens were incubated for 21 days at 25 °C in the dark, then oven dried for 48 hours at 60 °C. The dried specimens were stored at 4 °C and 65 % RH prior to testing. No mycelium was cultivated on the external boundaries of the specimens. No additive was used. The same cultivation protocol was used for all tests (compression, flexion, tension).

3.2.2 Mogu cultivation method

Materials

A *G. resinaceum* species from Mogu, with internal record 19-18, was used (GR_m). Hemp shives (cellulose: 35 %; hemicellulose: 18 %; lignin: 21 %; protein, pectin and other products: 18 %) were acquired from HempFlax BV. (ref. 7000277; Oude Pekela, The Netherlands). Coffee ground was acquired from a grocery store local to Mogu facilities, filtered for 5 min with boiling water, and the spent coffee ground was collected for use as an additive.

Protocol

Mogu manufactured a series of specimens for flexural characterisation. The principal substrates, fibres, and hessian were prepared at 60 % MC with mineralized water, and sterilised at 121 °C for 90 min. The substrates were then mixed with 3 wt% spawn and incubated for 21 days at 25 °C in the dark. Once colonised, the principal substrates were broken down and formed with the sterile fibre and hessian into aerated moulds. The formed specimens were incubated for 7 days at 25 °C in the dark, then oven dried for 72 hours at 70 °C.

3.2.3 Chemical analysis

Fourier-Transform Infrared (FTIR) spectrometry has been used previously for analysing the lignocellulosic profiles of substrates and their relation to fungal degradation patterns (Pandey and Pitman 2003; Pandey 1999; Haneef *et al.* 2017; Elsacker *et al.* 2019), with the benefit of requiring a limited specimen preparation, and spectra shape and frequencies being directly related to microscopical physical quantities and hence prepared for interpretation (Grdadolnik 2002). FTIR spectrometry was conducted in this study on a single reflection diamond Attenuated Total Reflectance (ATR) Agilent 4500a FTIR (Santa Clara, USA). The acquisition resolution was 4 cm^{-1} with 16 scans per specimen, for a band between 4000 cm^{-1} and 650 cm^{-1}. The baseline of FTIR spectra was corrected following the adaptive iteratively reweighted Penalised Least Squares (airPLS) method (Z.-M. Zhang *et al.* 2010), and spectra normalisation was done with amide I/II band envelopes (Baker *et al.* 2014). Four samples were isolated from *G. lucidum* colonised beech wood specimens, their spectra were averaged for analysis. The other materials were scanned with one replicate. Chemical analysis of *G. lucidum* colonised beech was performed on specimens having been tested in compression previously.

3.2.4 Compression series

Following ASTM D1037, compression parallel to surface evaluation is reported, for which the short-column method has been chosen as the specimens have a nominal thickness > 25 mm. They are parallelepipeds of 1:1:4 ratio, with dry dimension of 34 x 34 x 140 mm.

Effect of particle size & reinforcements

Four specimen groups were designed:

- Principal substrate as small granulation beech wood,
- Principal substrate as medium granulation beech wood,
- Principal substrate as large granulation beech wood,
- Principal substrate as mixed granulation beech wood,

Additionally, macro composition strategies were investigated in four groups:

- Control: principal substrate only,
- Hessian jacketing: a hessian jacketing was introduced in the length,
- Fibres perpendicular to load: 32 mm long rattan fibres were positioned regularly within the principal substrate as two layers of fibres, centred in the specimen thickness and separated by a 10 mm layer of principal substrate,
- Fibres coaxial to load: eight to ten fibres of common reed of 1 mm ± 0.5 diameter were chosen so as to balance their dimensional variability and positioned in two layers separated by 10 mm of principal substrate.

No external mycelium was left to grow on the external boundaries of the specimens so as to observe the effect of experimental variables without introducing a specimen geometry bias. This bias may be important for the reproducibility of experiments as the characteristics of the external mycelium mat are never found to be reported in the state-of-the-art. A jacketing strategy has been integrated to study the effect of boundary reinforcement. Six replicates were produced and tested for each specimen type. Load testing was performed on a Mecmesin MultiTest-dV testing bench equipped with a 2500 N load sensor, with a loading speed of 1.0 mm/min. Compressive modulus and ultimate compressive strength were calculated following ASTM D1037.

Effect of species and lignin

A medium granulation of beech wood cultivated *G. resinaceum* was tested with an identical cultivation protocol (BM_GR group). Pine wood sawdust was collected from a milling workshop and prepared following the previously described protocol with a *G. lucidum* species. These results are discussed with regards to *G. lucidum* grown medium beech wood (BM). 5 replicates have been tested for the BM_GR group, and only two could be tested for the PM_GL group. Load testing was performed on a Mecmesin MultiTest-dV testing bench equipped with a 2500 N load sensor. These specimens were tested at a loading speed of 4.0 mm/min. Young's modulus and ultimate compressive strength were calculated following ASTM D1037.

3.2.5 Flexion series

The experimental plan is designed to investigate the effect of reinforcement strategies over the flexural modulus and ultimate flexural strength. Following ASTM D1037, three point bending is reported. To this end, four specimen groups were designed:

- Control: no fibre,
- Inner hessian: a flat layer of hessian was introduced at mid-thickness,
- Hessian jacketing: a hessian jacketing was introduced in the length,
- Rattan: five parallel rattan fibres of 6 mm diameter by 500 mm, separated by 8 mm, were introduced in the length and at mid-thickness.

Six replicates were produced and tested for each of the specimen groups. Load testing was performed on a Mecmesin MultiTest-dV testing bench equipped with a 2500 N load sensor, with a loading speed of 10.0 mm/min. Flexural modulus and ultimate flexural strength were calculated following ASTM D1037. Two specimen series were produced, one by CITA following the protocol in section 3.2.1, and one by Mogu following the protocol in section 3.2.2. The wet specimens cultivated by CITA are parallelepipeds of 520 x 72 mm, with a nominal thickness of 20 mm. The width and thickness were not affected by the desiccation, but the length of the dry specimens varied between reinforcement strategies and shrank on average by 3.5 % in the control group, 2.7 % in the inner hessian group, 1.7 % in the hessian jacketing group, and 0.8 % in the rattan group. Mogu cultivated specimens exhibit a volumetric variability of up to 23.8 %. Calculations are taking the dimensional variability into account as averaged per specimen group. Detail pictures of a comparable specimen of each series are presented in Fig.3.21 and Fig.3.22.

3.2.6 Tension series

Following ASTM D1037, dog-bone specimens of a nominal thickness of 20 mm were cultivated. Tension parallel to surface is reported. Our experimental plan investigates the effect of granulate sizes over the tensile modulus and ultimate tensile strength. To this end, two specimen groups were designed:

- Small granulation of beech wood,
- Medium granulation of beech wood.

Six replicates were produced and tested for each of the specimen types. Load testing was performed on a Mecmesin MultiTest-dV testing bench equipped with a 2500 N load sensor, with a loading speed of 4.0 mm/min. Tensile modulus and ultimate tensile strength were calculated following ASTM D1037.

Figure 3.21: Detail in top view of CITA cultivated specimen (left), and Mogu cultivated specimen (right) (2021).

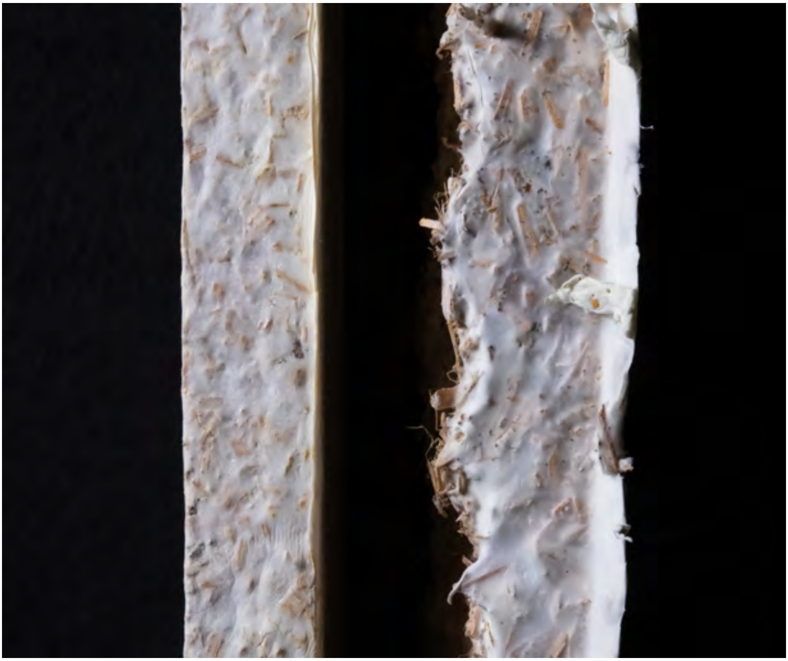

Figure 3.22: Detail in side view of CITA cultivated specimen (left), and Mogu cultivated specimen (right) (2021).

3.3 Chemical characterisation

FTIR spectrometry was used to characterise the four raw materials used in the compression series investigating composition strategies (section 3.2.1). The materials were hessian, beech wood, rattan and common reed. Their spectra are presented on Fig.3.23. Beech wood and rattan spectra

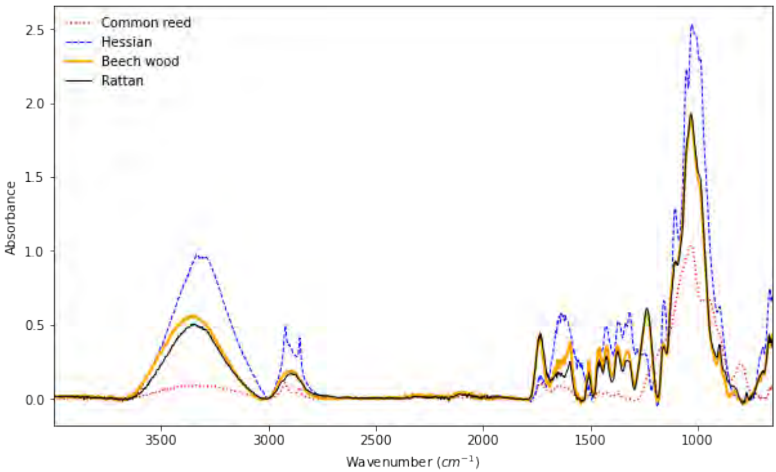

Figure 3.23: FTIR spectra of hemp-based hessian, beech wood, rattan, and common reed fibres (2021).

display a chemical profile that is very similar, with the exception of peaks at 1123 cm^{-1} and 1160 cm^{-1}, and the 1300 – 1500 cm^{-1} region. This indicates a slightly higher content of cellulose, hemicellulose and lignin in our tested beech wood specimen (C–O stretching, C–O–C asymmetrical stretching, C–H deformation, COOH groups symmetrical stretching, symmetric C–H bending, CH_2 deformation stretching, CH_3 asymmetrical angular vibration, vibrational mode of amide C–O stretching) (Bekiaris *et al.* 2020;

X. Li *et al.* 2018; Shi *et al.* 2012). Common reed displays a minimal amount of lignin and hemicellulose compared to our other samples, while the peak at 890 cm^{-1} is associated with C–O–C stretching at the *β*-(1→4)-glycosidic linkages of amorphous cellulose (Ciolacu *et al.* 2011). The hessian displays distinctive peaks at 707 cm^{-1}, 890 cm^{-1}, 1060 cm^{-1}, 1316 cm^{-1}, and 1430 cm^{-1} in the fingerprint region, and 1640 cm^{-1}, and 2921 cm^{-1}. The 750 – 680 cm^{-1} and 1680 – 1630 cm^{-1} regions (C=O streching) are associated with primary and secondary amides in hemp (amide V: C–N and N–H vibrations) (Smith 2020). Primary amides in hemp are amino acids, fatty acids, and steroids, which contribute to the 3500 – 3000 cm^{-1} region. The 1310 – 1230 cm^{-1} region (C–N stretching) is associated to secondary amides, such as cannabinoids, flavonoids, stilbenoids, terpenoids, alkaloids, and lignans (Flores-Sanchez and Verpoorte 2008). The peak at 2921 cm^{-1} is associated with alkyl C–H groups (Dorado *et al.* 2001).
Four samples were isolated from *G. lucidum* colonised beech wood specimens after they were used for load testing. Their spectra were averaged and are presented on Fig.3.24 along with the beech wood spectrum, and a sample scan of *G. lucidum* mycelium. Peaks at 886 cm^{-1}, 1075 cm^{-1} and 1160 cm^{-1} are characteristic of (1→3)- and (1→6)-*β*-glucans that are present in the fungal cell wall (identified as [2], and [4] on Fig.3.24). The peaks [1] and [3] at 780 cm^{-1} and 1043 cm^{-1} are also associated with *β*-glucans (Šandula *et al.* 1999). Chitin is identified at peak [5] 1313 cm^{-1} (amide III: C–N stretching), which also affects the 1640 cm^{-1} region [6] alongside the presence of peptides and secondary metabolites (aromatic rings and conjugated alkenes). The peak [7] at 2922 cm^{-1} is representative of chitin and ergosterol (C–H stretching) (Girometta *et al.* 2019). The 3600 – 3000 cm^{-1} region (peak [8]) is considered to be influenced by residual water and entrapped CO_2 (O–H and N–H stretching). Finally, peaks [9] to [12] represent decreases at 1231 cm^{-1}, 1425 cm^{-1}, 1506 cm^{-1}, and 1733 cm^{-1}. They are associated with lignin and xylan breakdown (syringyl ring breathing and C–O stretching,

C=C stretching vibration in aromatic ring), and cellulose (peak [11]) and hemicellulose (peak [11] and [12]) breakdown is observed (CH_2 scissor vibration, C=O stretching) (Cai *et al.* 2018). To evaluate the lignocellulosic changes undertaken during *G. lucidum* activity quantitatively, the band ratio indices at 1231 cm^{-1}, 1425 cm^{-1}, and 1506 cm^{-1} were calculated from the 2921 cm^{-1} band (Diehl *et al.* 2003) for beech wood and *G. lucidum* colonised beech wood as:

$$\frac{I_n}{I_{2921}}, \tag{3.2}$$

Where I_n is the specific band intensity and I_{2921} the band intensity at 2921 cm^{-1}. The band ratio at 1231 cm^{-1} went from 1.58 in beech wood to 0.61 in *G. lucidum* colonised beech wood; the band ratio at 1425 cm^{-1} went from 0.96 in beech wood to 0.63 in *G. lucidum* colonised beech wood; the band ratio at 1506 cm^{-1} went from 0.71 in beech wood to 0.24 in *G. lucidum* colonised beech wood; the band ratio at 1733 cm^{-1} went from 1.19 in beech wood to 0.41 in *G. lucidum* colonised beech wood. It can therefore be observed that *G. lucidum* had a preference in breaking down lignin and xylan at 1231 cm^{-1} compared to cellulose and hemicellulose at 1425 cm^{-1} (2.94:1), which is confirmed by the ratios at 1506 cm^{-1} for lignin (1.42:1), and 1733 cm^{-1} for hemicellulose (2.36:1). The CH_2 scissor vibration corresponding to the peak at 1425 cm^{-1} reflecting both cellulose and hemicellulose, the present decrease might be primarily related to hemicellulose breakdown. This preference of *G. lucidum* for lignin and hemicellulose is consistent with findings reported in the literature (Zhou *et al.* 2018).

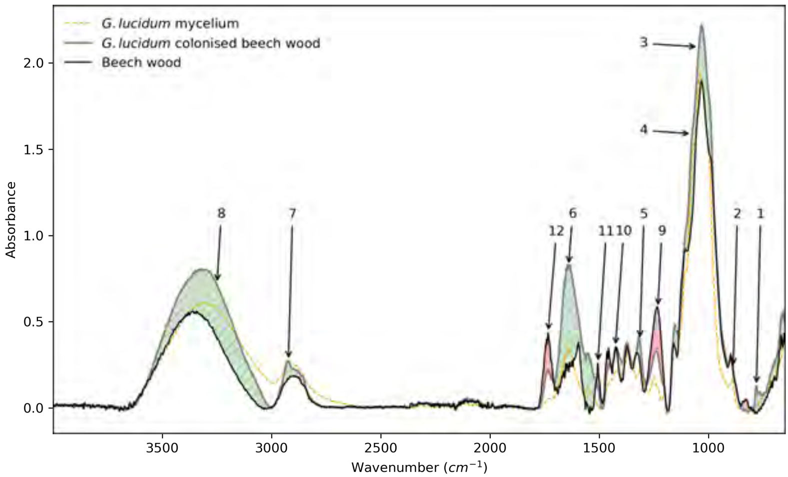

Figure 3.24: FTIR spectra of *G. lucidum* colonised beech wood, *G. lucidum* mycelium, and beech wood. Green areas represent increased values in mycelium-colonised specimens (peaks 1 to 8), red areas are decreased values in mycelium-colonised specimens (peaks 9 to 12) (2021).

3.4 Mechanical characterisation

3.4.1 Effect of particle size & composition in compression

The effect of particle sizes on the mechanical behaviour in compression of mycobosci was investigated using four levels of granulation: small (BS family), medium (BM family), large particles (BL family), and a 1:1:1 volume ratio mix of the three previous granulations (BSML family). A second parameter was introduced to investigate the anisotropic modification of mycobosci. Three typologies of fibre composition were implemented in the experimental plan: hessian jacketing coaxial to the load case (H), unidirectional rattan fibres perpendicular to the load case (R), and unidirectional common reed fibres coaxial to the load case (V). Isotropic controls were added for each level of granulation (BS, BM, BL, BSML specimen types in the figures). Fig.3.25 illustrates the three typologies alongside the control. Experimental parameters per specimen type and resulting mean density, mean Young's modulus and mean ultimate strength are presented in table 3.11. Box plots of the results for Young's modulus and ultimate strength are presented in Fig.3.26. A picture of a specimen from the series is presented in Fig.3.27, a group of specimens in Fig.3.28, and specimens after having been tested in Fig.3.30 and Fig.3.31.

Jacketing coaxial to load

The introduction of the hessian jacket offers a contrasting illustration of the effect of the mycelial mat usually grown on the external boundary of mycobosci. It can be observed that the dispersion of Young's modulus results across all specimen families is reduced compared to their controls with the exception of the BL family. The jacketing also affects the dispersion of results in ultimate strength in the case of the BS, BM, and BSML

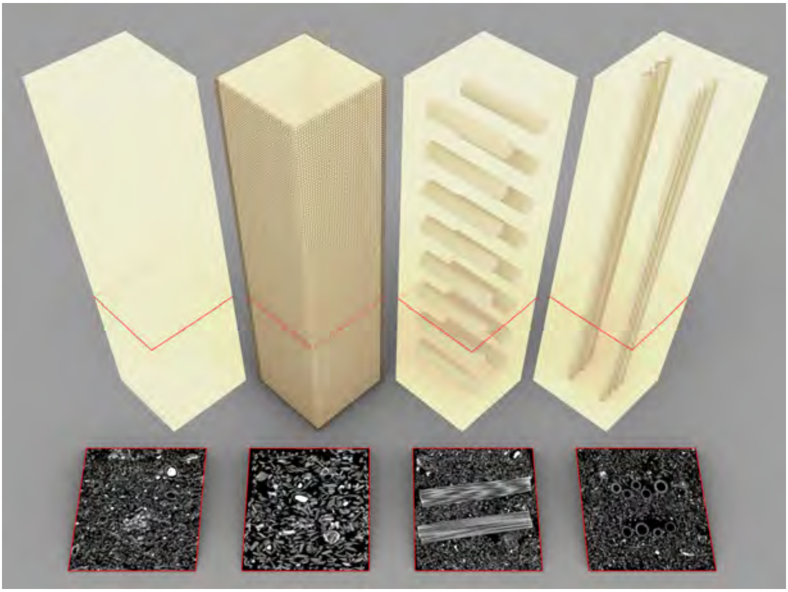

Figure 3.25: Fibre placement strategies and their sectional CT scan (left to right): control (BS), jacketing coaxial to load (BM_H), fibres perpendicular to load (BS_-R), fibres coaxial to load (BS_V).

Type	**Granulate size**	**Fibre composition**	**Mean density** (s.d.)	**Mean Young's modulus** (s.d.)	**Mean ultimate strength** (s.d.)
BS	0.5 – 1.0 mm	Control	209.67 kg/m^3 (6.47)	1.79 MPa (0.41)	171.86 kPa (36.54)
BS_H	0.5 – 1.0 mm	Hessian jacketing	230.48 kg/m^3 (9.88)	1.58 MPa (0.42)	175.79 kPa (34.38)
BS_R	0.5 – 1.0 mm	Rattan perpendicular	196.59 kg/m^3 (19.01)	0.66 MPa (0.42)	89.06 kPa (58.49)
BS_V	0.5 – 1.0 mm	Common reed coaxial	194.12 kg/m^3 (5.09)	3.88 MPa (2.51)	146.85 kPa (39.26)
BM	0.75 – 3.0 mm	Control	233.87 kg/m^3 (9.04)	3.32 MPa (0.80)	306.38 kPa (57.64)
BM_H	0.75 – 3.0 mm	Hessian jacketing	248.70 kg/m^3 (12.20)	2.99 MPa (0.54)	298.85 kPa (35.47)
BM_R	0.75 – 3.0 mm	Rattan perpendicular	226.77 kg/m^3 (6.19)	4.02 MPa (4.45)	232.30 kPa (62.85)
BM_V	0.75 – 3.0 mm	Common reed coaxial	198.14 kg/m^3 (2.89)	9.21 MPa (6.42)	270.93 kPa (79.76)
BL	4.0 – 12.0 mm	Control	217.60 kg/m^3 (10.58)	2.96 MPa (1.04)	245.60 kPa (30.31)
BL_H	4.0 – 12.0 mm	Hessian jacketing	264.05 kg/m^3 (11.97)	3.01 MPa (0.46)	223.93 kPa (25.88)
BL_R	4.0 – 12.0 mm	Rattan perpendicular	240.98 kg/m^3 (3.91)	2.24 MPa (0.58)	180.88 kPa (64.75)
BL_V	4.0 – 12.0 mm	Common reed coaxial	209.47 kg/m^3 (7.90)	8.50 MPa (4.56)	290.86 kPa (100.83)
BSML	0.5 – 12.0 mm	Control	220.59 kg/m^3 (8.12)	2.17 MPa (0.36)	237.09 kPa (31.73)
BSML_H	0.5 – 12.0 mm	Hessian jacketing	246.85 kg/m^3 (11.29)	2.20 MPa (1.04)	194.48 kPa (48.47)
BSML_R	0.5 – 12.0 mm	Rattan perpendicular	224.09 kg/m^3 (3.55)	1.87 MPa (0.30)	171.44 kPa (26.13)
BSML_V	0.5 – 12.0 mm	Common reed coaxial	203.08 kg/m^3 (6.24)	7.89 MPa (2.41)	338.75 kPa (65.39)

Table 3.11: Summary of specimen types parameters, resulting dried densities, and compressive properties.

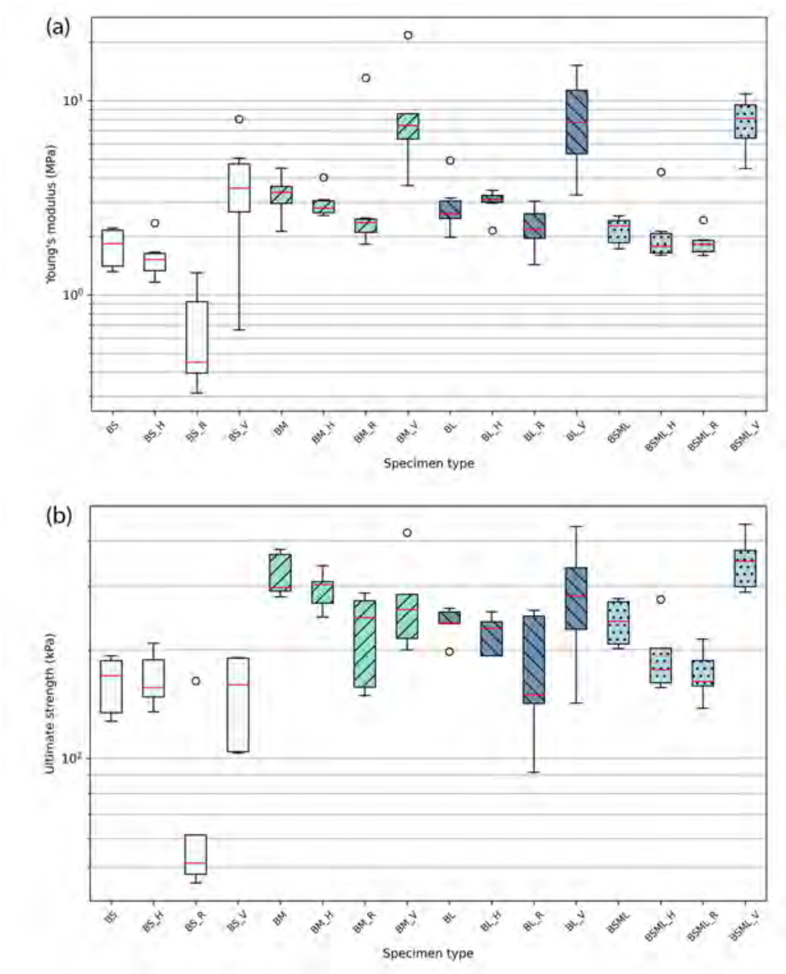

Figure 3.26: Box plots for compressive Young's modulus results (a) and ultimate strength results (b).

Figure 3.27: One dried specimen from the compression series.

Figure 3.28: Specimens from the compression series.

families, with a reduction of the deviation between the first and third quartiles. The containment of stress applied to the specimens within tight boundaries forces the arrangement of the particles within, restricting the ability for particles to arrange freely. Jacketed specimens have an average reduction of 0.12 MPa to the controls as per Young's modulus (s.d. 0.19), and an average decrease of 16.97 kPa to the controls as per ultimate strength (s.d. 20.05). The jacket has two important advantages: it offers a durable alternative to low-ductility mycelial mats usually grown on the external boundary of mycobosci, and it can be hypothesised that it would substantially contribute to an increase in fracture resistance performance in shearing and bending load cases.

Fibres perpendicular to load

Specimens supplemented with rattan fibres display a lower performance across particle sizes considering their median in Young's modulus and ultimate strength. The mean ultimate strength follows the performance of the mean of the controls (Fig.3.29) with an average reduction of 71.81 kPa (s.d. 8.45). This suggests that, should the production conditions of such mycoboscus improve to reduce the dispersion of results and increase the material behaviour predictability, introducing strategically parsed weakness points in composites could find a use with calibrated materials by tuning their failure mode.

Fibres coaxial to load

Common reed fibre reinforced specimens resulted in the largest standard deviations in Young's moduli (reported in table 3.11), especially in the BM and BL families. This is due to the fibres having partially misaligned to the load case axis during specimen production. Nevertheless, results suggest that mycobosci can be successfully stiffened with regards to their

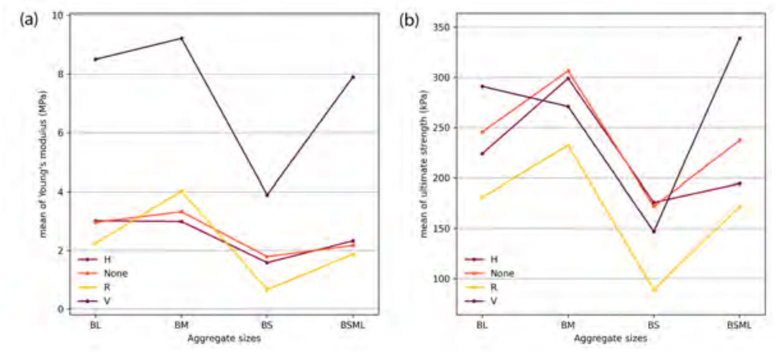

Figure 3.29: Parameters interaction graph for mean Young's modulus (a) and mean ultimate strength (b).

use case. The effect of this stiffening on the ultimate strength is less obvious as it can be noted that the smaller particles (BS and BM families) tend to perform better without reinforcement coaxial to load. This is a result of the inherent large displacement of the fibres within the specimens under stress due to their stiffness, thus initiating an early critical failure. The mean Young's moduli (table 3.11) display a clear improvement compared to the controls: an average increase of a factor 2.86 (s.d. 0.6) is observed between the mean of the controls and the mean of the fibre coaxial to load specimens. As per mean ultimate strengths, they improved in the BL and BSML families when compared to controls (respectively by a factor 1.18 and 1.43), but decreased in the smaller particles families BS and BM (respectively by a factor 0.86 and 0.88).

Principal substrate particles

The use of smaller particles in mycobosci increases the specific surface of the substrate, hence facilitating the access to nutrients and initial air distribution. The best performing particle size is BM (0.75 – 3.0 mm) and not BS (0.5 – 1.0 mm). This can be explained by the gas permeability of the substrate and its influence on local CO_2 build ups resulting from oxidative decay, itself influencing the rate of decomposition of the particles. An analysis of the phenomena at play with regards to particle size is presented in section 3.4.5 and in the conclusions of the chapter (section 3.5).

Statistical analysis

The result distributions are two-tailed. The mean of Fisher's defined kurtosis for Young's modulus series is −0.3328 (s.d. 0.9353) and −1.0564 for ultimate strength (s.d. 0.5343). Fisher-Pearson's skewness coefficient mean for Young's modulus is 0.5834 (s.d. 0.8540), and 0.1733 for ultimate strength (s.d. 0.5137). The distributions are considered normal (Hair *et al.* 2013), which was verified for ultimate strength and Young's modulus results with the Shapiro-Wilk test (respectively p=0.9224 and p=0.0030, α=0.001). Equality of variances was controlled with the Levene test; Young's modulus result variances are not equal (p=1.3940×10^{-5}, α=0.05), neither are ultimate strength ones (p=0.0459, α=0.05). Welch's ANOVA was conducted for the two parameters: fibre placement for Young's modulus and ultimate strength (respectively p=0.0001 and p=0.0013), and particle size for Young's modulus and ultimate strength (respectively p=0.0030 and p=4.6462×10^{-9}). The mean values of specimen groups are significantly different (α=0.005). Using the pairwise Games-Howell test the most significant reinforcement is identified to be the fibre coaxial to load against fibre perpendicular to load, the control, and hessian jacketing (all p=0.001 as per Young's modulus; respectively p=0.030, p=0.004, and p=0.004 as per ultimate strength; α=0.05). Continuing this test, the most significant aggregate size is identified to be the 0.5 – 1.0 mm interval (BS family) against the BM and BL families (respectively p=0.029 and p=0.036 as per Young's modulus; all p=0.001 as per ultimate strength;

α=0.05). The BS family had a significant difference to the BSML family as per aggregate size over ultimate strength (p=0.001, α=0.05), but not over Young's modulus (p=0.106, α=0.05).

3.4.2 Effect of particle size in tension

The effect of particle size on the mechanical behaviour in tension of mycobosci was investigated using two granulations: small (BS) and medium (BM). The dog-bone specimen design is presented in Fig.3.32; the smallest section of the dried dog-bones was of 20 x 36 mm, and 51 mm in length. Experimental parameters per specimen type and resulting mean density, mean Young's modulus and mean ultimate strength, are presented in table 3.12. Box plots of the results for Young's modulus and ultimate strength are presented in Fig.3.34. While the difference in dried composite density between the two groups was of 1.17-fold, the stiffness increase from BS to BM specimens was of 1.53-fold. An analysis of the phenomena at play with regards to particle size is presented in section 3.4.5 and in the conclusions of the chapter (section 3.5).

Statistical analysis

The result distributions are two-tailed. The mean of Fisher's defined kurtosis for Young's modulus series is –1.4313 (s.d. 0.0889) and –1.4840 for ultimate strength (s.d. 0.0013). Fisher-Pearson's skewness coefficient mean for Young's modulus is –0.2065 (s.d. 0.4445), and –0.1546 for ultimate strength (s.d. 0.5246). The distributions are considered normal (Hair *et al.* 2013), which was verified for ultimate strength and Young's modulus results with the Shapiro-Wilk test (α=0.05). Equality of variances was controlled with the Levene test; Young's modulus and ultimate strength variances are considered equal (respectively p=0.1011, p=0.7427; α=0.05). Welch's ANOVA was conducted for Young's modulus and ulti-

Figure 3.30: Rattan embedding specimen used for compressive characterisation, after having been tested.

Figure 3.31: Hessian jacketed specimen used for compressive characterisation, after having been tested.

Type	Granulate size	**Mean density** (s.d.)	**Mean Young's modulus** (s.d.)	**Mean ultimate strength** (s.d.)
BS	0.5 – 1.0 mm	185.15 kg/m^3 (4.82)	0.55 MPa (0.08)	0.07 MPa (0.01)
BM	0.75 – 3.0 mm	217.35 kg/m^3 (17.71)	0.84 MPa (0.15)	0.08 MPa (0.01)

Table 3.12: Summary of specimen types parameters, resulting dried densities, and tensile properties.

mate strength (respectively p=0.0030 and p=0.0238). The mean values of specimen groups are significantly different (α=0.05). Using the pairwise Games-Howell test the significant difference between groups was confirmed (α=0.05).

3.4.3 Effect of composition in flexion

The effect of diverse reinforcements on the mechanical behaviour in flexion of mycobosci was tested, using three levels: inner hessian (I), hessian jacketing (H), and rattan fibres in the length (R). These strategies are illustrated in Fig.3.35, and a picture of specimens is presented in Fig.3.36. The principal substrate of the specimens is the medium granulation of beech wood (0.75 – 3.0 mm). The BM series was made following the standard protocol (section 3.2.1), and the HC series was made following protocol Mogu (section 3.2.2). Isotropic controls were added to the experimental series (BM and HC groups). Experimental parameters per specimen type and resulting mean density, mean Young's modulus and mean modulus of rupture are presented in table 3.13. Box plots of the results for Young's modulus and modulus of rupture are presented in Fig.3.37, and parameters interaction in Fig.3.38.

It can be noted that the mechanical failure of the specimens was related to dewetting of the principal substrate across all groups and at the level of the highest tensile stress, that is at the middle of the span on the opposite surface to the applied load. Rattan fibres did not fail nor deform

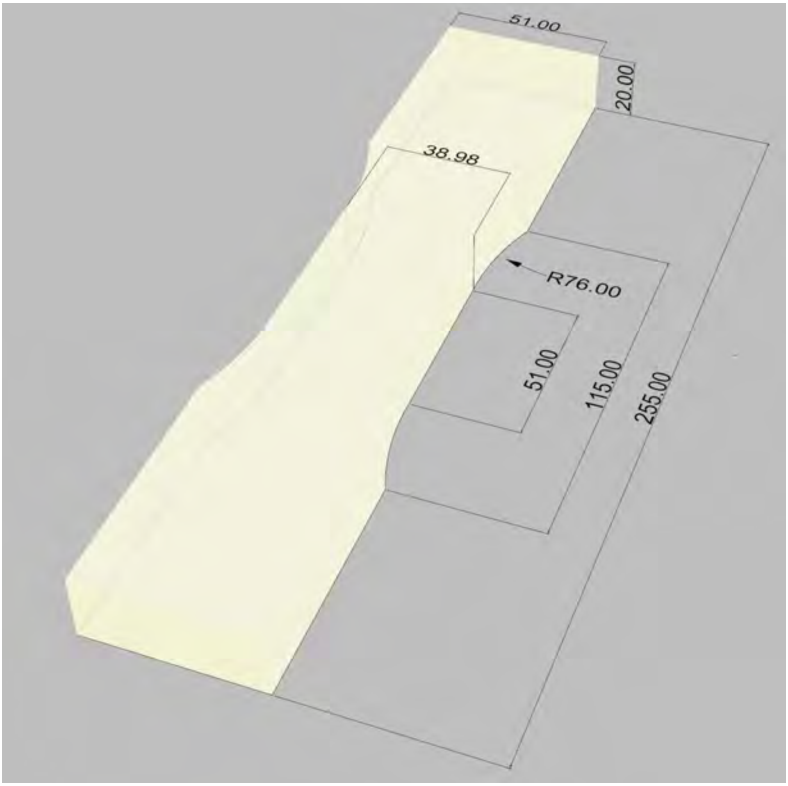

Figure 3.32: ASTM D1037 informed dog-bone design for the flexion series (wet specimens dimensions).

Figure 3.33: Two of the specimens tested for tensile characterisation (BM and BS-based); notice the arrangement of particles and mycelium in the section.

plastically. Likewise, no debonding of the hessian jacketing was visible after testing. Representative failure modes for the BM series are presented in Fig.3.39. Limited fractures in the most elastic specimen groups BM and BM_I can be appreciated. This is where the mycelial matrix also deformed plastically on the surface most exposed to tensile stress (10 mm from the neutral axis). A lack of external fracture in the BM_H specimen group can be observed. The BM_R group resulted in more external fractures as the aligned continuous rattan fibres were favourable to increasing strength and stiffness while its continuous interfacial bond between the matrix and the rattan fibres constrained the deformation. A future investigation may focus on displacing the fibre reinforcement towards the most stressed opposite surface to the load, distancing it from the neutral axis to improve its efficiency. A backdrop that could be expected from this is the earlier formation of fractures on the surface exposed to tensile

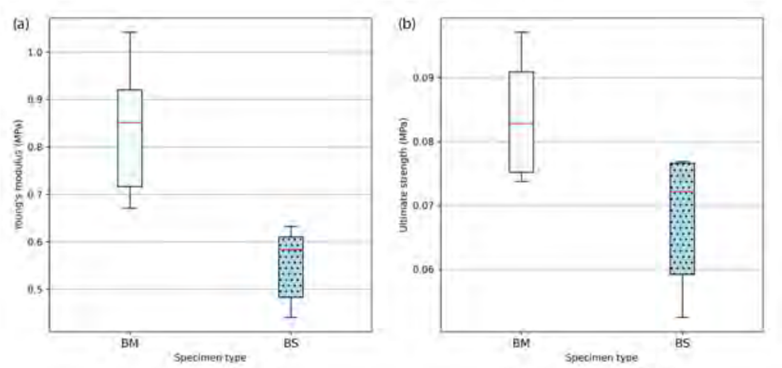

Figure 3.34: Box plots for Young's modulus results (a) and ultimate tensile strength results (b).

stress, while the fibre alignment with the neutral axis in this experimental series left a thicker sectional area of beech wood and mycelium complex under the fibres, which allowed for the mycelial matrix to deform elastically and plastically to a greater strain. In Fig.3.36 tested specimens crossing at mid-length can be observed: they have been softened by aggregates debonding but stayed integral.

Similarly to the investigation on compression behaviour characterisation, the hessian jacketing series offers a contrasting instance of the effect that the cultivation of an external mycelial skin on test specimen might lead to. The BM_H series resulted in a 1.95-fold increase in flexural modulus as compared to the control group, and 1.5-fold increase in strength. In contrast to the BM_I series which had a hessian reinforcement added at mid-thickness and along the neutral stress axis, jacketing is a particularly suitable fibre composition strategy as the low elasticity of hessian contributes to resisting the tensile stress on the surface opposite to the load and on covered side surfaces. Beech wood particles further contribute to the composite strength where subjected to compression—on the surface where the load is applied—as the elastic mycelial matrix reaches its maximal strain and beech particles of a higher stiffness interlock. The BM_R series has the highest relative standard deviation with 41.3 % of the mean, while other groups have a standard deviation of a maximum of 27.2 %. An extremum is reported at 2.44 GPa while Q3 is 1.35 GPa for a mean at 1.34 GPa. If not considering this extremum, the mean of the series is at 1.12 GPa for a standard deviation closer to mycobosci standards at 211.06 MPa (18.8 %).

Statistical analysis

The result distributions are two-tailed. The mean of Fisher's defined kurtosis for Young's modulus series is −0.8114 (s.d. 0.5732) and −0.5973 for modulus of rupture (s.d. 0.3400). Fisher-Pearson's skewness coefficient mean for Young's modulus is 0.4158 (s.d. 0.6169), and −0.1262 for modulus of rupture (s.d. 0.7371). The distributions are considered normal (Hair *et al.* 2013), but did not satisfy the Shapiro-Wilk test (modulus: $p=2.1097 \times 10^{-9}$, modulus of rupture: $p=2.7600 \times 10^{-7}$, $\alpha=0.01$). Equality of variances was controlled with the Levene test; Young's modulus result variances are not equal ($p=0.0005$, $\alpha=0.05$), neither are modulus of rupture ones ($p=9.9545 \times 10^{-5}$, $\alpha=0.05$). Welch's ANOVA was conducted for Young's modulus and modulus of rupture regarding reinforcement strategies (respectively $p=0.035$ and $p=0.000009$), and regarding "cultivation media", between CITA and Mogu protocols (respectively $p=0.0071$ and $p=0.043$). The mean values are significantly different ($\alpha=0.05$). Across the two series, using the pairwise Games-Howell test the significant difference between the rattan group and the other reinforcements for Young's modulus and modulus of rupture was confirmed ($\alpha=0.05$), besides for inner hessian group against the controls, and hessian jacketing against rattan ($p=0.062$). The significant difference between cultivation

Type	Fibre composition	**Mean density** (s.d.)	**Mean Young's modulus** (s.d.)	**Mean modulus of rupture** (s.d.)
BM	Control	216.27 kg/m^3 (17.00)	192.71 MPa (52.40)	0.12 MPa (0.03)
BM_I	Inner hessian	215.12 kg/m^3 (8.01)	197.33 MPa (45.56)	0.11 MPa (0.02)
BM_H	Hessian jacketing	228.77 kg/m^3 (11.60)	375.14 MPa (1.82)	0.18 MPa (0.03)
BM_R	Rattan	245.51 kg/m^3 (9.62)	1.38×10^3 MPa (630.13)	0.62 MPa (0.14)
HC	Control	123.55 kg/m^3 (2.42)	38.30 MPa (6.72)	0.05 MPa (0.01)
HC_I	Inner hessian	94.74 kg/m^3 (10.96)	20.41 MPa (11.48)	0.04 MPa (0.02)
HC_H	Hessian jacketing	93.49 kg/m^3 (4.98)	11.02 MPa (3.73)	0.03 MPa (0.01)
HC_R	Rattan	103.51 kg/m^3 (6.11)	318.19 MPa (68.50)	0.31 MPa (0.02)

Table 3.13: Summary of specimen types parameters, resulting dried densities, and flexural properties.

media for Young's modulus and modulus of rupture was confirmed too (α=0.02). In the CITA cultivated series the significant difference between all groups for Young's modulus and modulus of rupture was confirmed (α=0.05), but for inner hessian against control.

3.4.4 Effect of species and lignin

The effect of species on the mechanical behaviour in compression of mycobosci was investigated with a *G. resinaceum* cultivated on medium granulate size beech wood (Fig.3.42), and comparing results to the BM group of the experimental series presented in section 3.4.1. Five replicates could be tested in this group. So as to confirm the lower softwood decaying potential of the *G. lucidum* species, a pine wood specimen group was also tested. Because of a low colonisation ratio as compared to hardwood cultivated composites, only two replicates could be tested for this configuration (Fig.3.41). Even with a lower number of replicates, it appears that the pine substrate is not a relevant substrate for the *G. lucidum* species with the cultivation protocol that was used. This limited screening is nonetheless consistent with section 2.9 regarding guaiacyl lignin presence in gymnospermous wood and their higher resistance to fungal decay. The poor

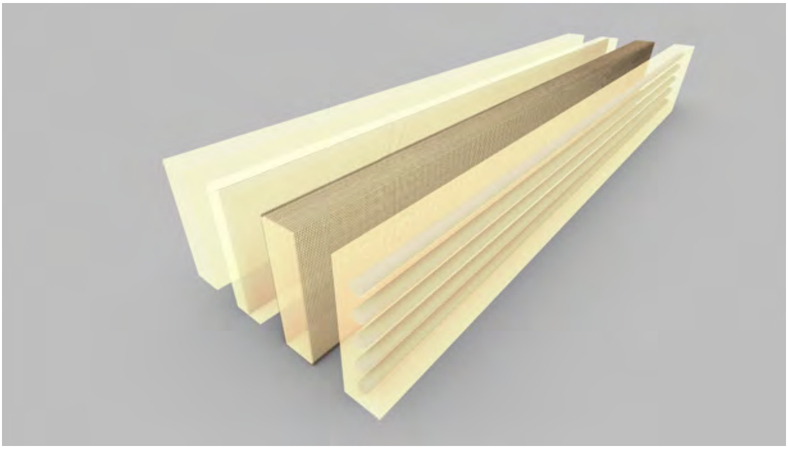

Figure 3.35: Fibre placement strategies in the flexion series (left to right): control, inner hessian, hessian jacketing, rattan fibres.

colonisation of PM_GL specimens is illustrated on Fig.3.41. *G. resinaceum* cultivated specimens resulted in a compressive behaviour similar to small granulation beech wood colonised by *G. lucidum*. The cultivation protocols were identical between these two series. Experimental parameters per specimen type and resulting mean density, mean Young's modulus and mean ultimate strength are presented in table 3.14. Box plots of the results for Young's modulus and ultimate strength are presented in Fig.3.40.

Statistical analysis

Because only two replicates could be tested for PM_GL, this series results are not considered significant and were not statistically analysed. The BM_GR series was tested against the BM series from section 3.4.1. The result distributions are two-tailed. The mean of Fisher's defined kurtosis for BM_GR Young's modulus is −1.8933 and −1.3775 for ultimate strength.

Figure 3.36: Standard cultivated specimens used for flexural characterisation, after having been tested.

Fisher-Pearson's skewness coefficient mean for BM_GR Young's modulus is 0.0806, and 0.4800 for ultimate strength. The distributions are considered normal (Hair *et al.* 2013), which was verified for ultimate strength and Young's modulus results with the Shapiro-Wilk test (respectively: $p=0.6605$, $p=0.9611$; $\alpha=0.5$). Equality of variances between the BM_GR and BM groups was controlled with the Levene test; Young's modulus and ultimate strength results variances are considered equal (respectively $p=0.0627$, $p=0.0799$; $\alpha=0.05$). Welch's ANOVA was conducted for Young's modulus and ultimate strength (respectively $p=0.0037$ and $p=0.0054$). The mean values of specimen groups are significantly different ($\alpha=0.01$). Using the pairwise Games-Howell test the significant difference between groups for Young's modulus and ultimate strength was confirmed (respectively $p=0.004$, $p=0.005$; $\alpha=0.005$).

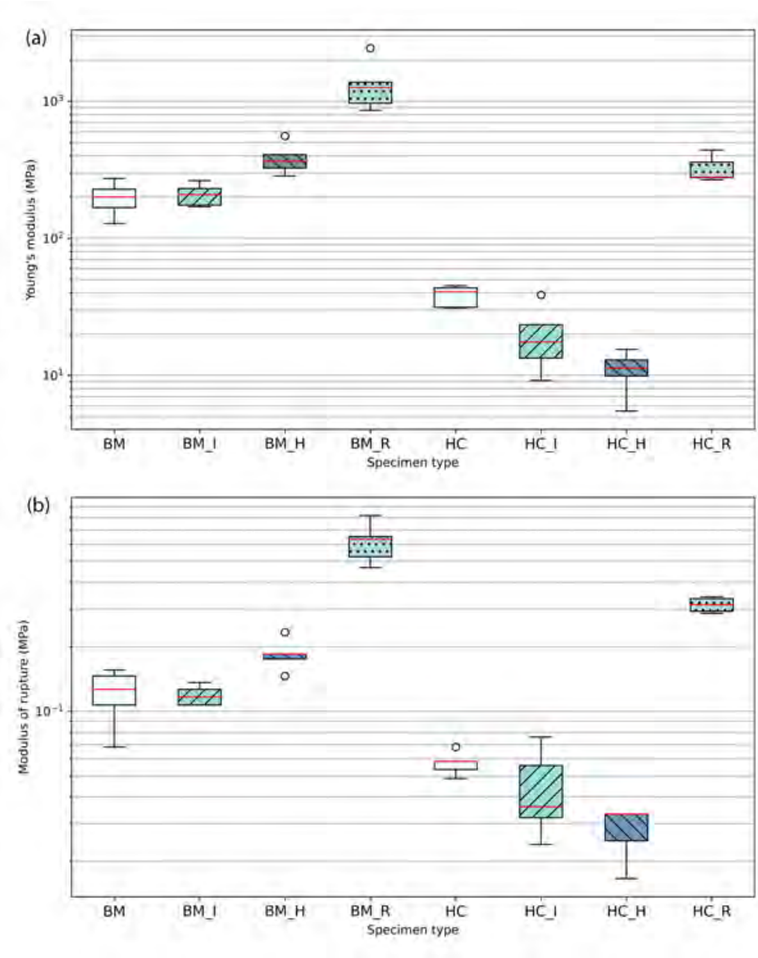

Figure 3.37: Box plots for Young's modulus results (a) and modulus of rupture results (b).

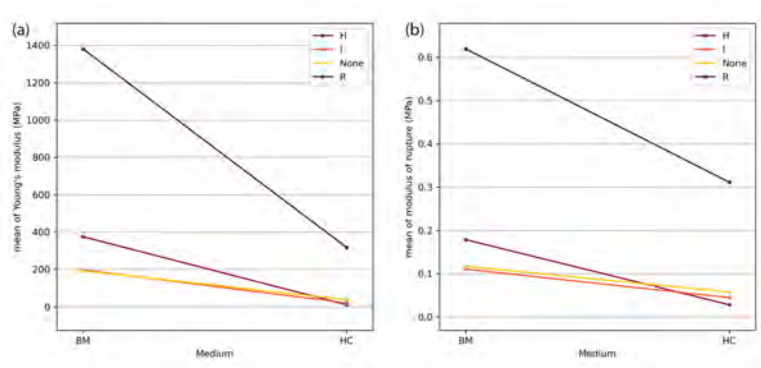

Figure 3.38: Parameters interaction graph for mean Young's modulus (a) and mean modulus of rupture (b).

3.4.5 Specific matrix modulus

The elastic modulus of a two phase particulate composite made of large particles can be predicted by the rule of mixture (Callister 1999 p.523). The upper bound representing axial loading for this rule is represented as:

$$E_{c+} = E_m \varphi_m + E_p \varphi_p \tag{3.3}$$

With E_m the Young's modulus of the matrix phase, φ_m the volume fraction of the matrix, E_p the Young's modulus of the dispersed phase (particles) and φ_p the volume fraction of the dispersed phase. The lower bound is given as:

$$E_{c-} = \frac{E_m E_p}{\varphi_m E_p + \varphi_p E_m} \tag{3.4}$$

This equation for the *inverse rule of mixture*, that represents transverse loading, does not take into consideration the matrix shielding from the aggregates and assumes equal stress distribution in the composite. This

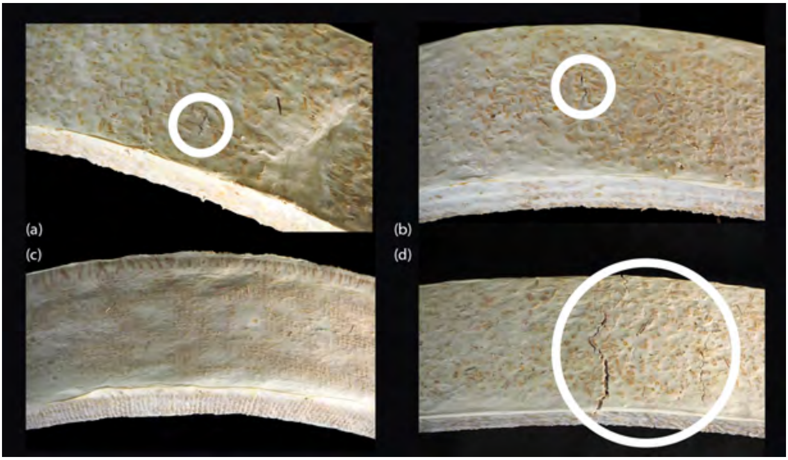

Figure 3.39: Representative failure modes for the (a) BM group, (b) BM_I group, (c) BM_H group, (d) and BM_R group.

makes it not entirely valid, although useful to approximate the system behaviour. The rule of mixture assumes a uniform distribution of fibres, a perfect bonding between fibres and the matrix, a homogeneous matrix, and a linear elastic stress response for the matrix and dispersed phase materials. The mechanical response of pure mycelium has been approached with a hyperelastic model (section 2.8; Islam *et al.* 2018a), and wood is a linear orthotropic viscoelastic material (Schniewind and Barrett 1972) which challenges the assumption of linear elastic stress response. The matrix is also full of voids. The theoretical volume fraction of mycelium is $\varphi_m = 1 - \varphi_p$, but as it retracts at hyphae level when dried and its grown density is heterogeneous in the volume given heterogeneous growth conditions, the exact volume can only be theoretically approximated here. Although the conditions are not met to apply the rule of mixture such that variable effects can be isolated, it was studied as a global method to evalu-

Type	Composition	*n*	**Mean density** (s.d.)	**Mean Young's modulus** (s.d.)	**Mean ultimate strength** (s.d.)
BM_GR	*G. resinaceum*, BM	5	207.49 kg/m^3 (5.79)	1.75 MPa (0.17)	0.11 MPa (0.02)
PM_GL	*G. lucidum*, pine wood	2	114.00 kg/m^3 (2.19)	0.34 MPa (0.00)	0.04 MPa (0.00)
BS	*G. lucidum*, BS	6	209.67 kg/m^3 (6.47)	1.79 MPa (0.41)	0.17 MPa (0.04)
BM	*G. lucidum*, BM	6	233.87 kg/m^3 (9.04)	3.32 MPa (0.80)	0.31 MPa (0.06)
BL	*G. lucidum*, BL	6	217.60 kg/m^3 (10.58)	2.96 MPa (1.04)	0.25 MPa (0.03)
BSML	*G. lucidum*, BSML	6	220.59 kg/m^3 (8.12)	2.17 MPa (0.36)	0.24 MPa (0.03)

Table 3.14: Summary of specimen types parameters, resulting dried densities, and compressive properties. Values in the lower section of the table are reported from section 3.4.1.

ate the mycoboscus quality and to extend the analysis of the experimental results.

In Fig.3.44 the effect of aeration on the grown mycelial density can be observed. This specimen was cultivated for four months with a *G. lucidum* species on spent substrates from the Empirical distillery. A 5 – 10 mm thick outer perimeter of the material is more fermented thanks to an increased air exchange (marked by the dashed curve). This can be observed with lignin degradation resulting in whiter wood particles in the area B, and with a denser mycelium. Conversely, in area A at the centre of the specimen the particles contain more lignin and look more brown, and the mycelium is less dense. This heterogeneity illustrates the effect of the build-up of CO_2 released with oxidative decay, and the fermentation activity is also influenced by the dynamic moisture conditions which are sensitive to the perimeter conditions. The ASTM-informed specimens geometries were not influenced by this gradient effect given their smaller dimensions. A section view of a compression specimen is presented in Fig.3.43 where the disposition of aggregates and mycelial matrix can be appreciated.

In spite of the departure from assumptions of the rule of mixture, the specific matrix modulus informs about the normalised binding performance as if the matrix phase was represented by a continuous binding agent.

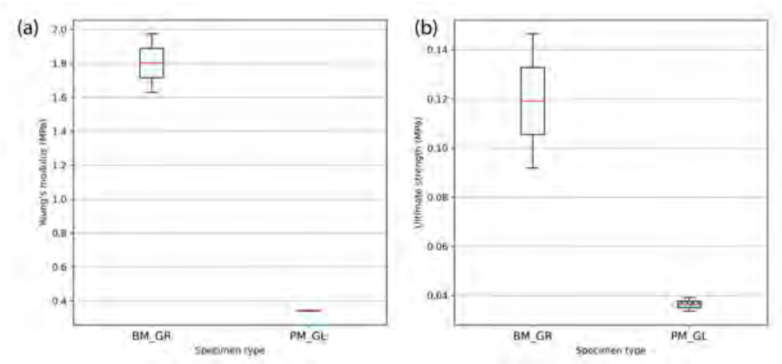

Figure 3.40: Box plots for Young's modulus results (a) and ultimate compressive strength results (b).

This metric is influenced by the following variables:

- Aggregate geometrical characteristics and distribution,
- Wood decay and softening,
- Mycelium hyperelastic behaviour,
- Fungal cell wall expression,
- Wood orthotropy,
- Discontinuity of the matrix phase,
- Heterogeneity of the matrix phase,
- Matrix shielding by the particles.

In the present case, the standardised supply of beech wood aggregates and absence of supplement use are hypothesised to lead to a similar fungal cell wall expression across specimen groups. Similarly, a standardised

Figure 3.41: One specimen of *G. lucidum* colonised pine wood used for compressive characterisation, after having been tested.

Figure 3.42: One specimen of *G. resinaceum* colonised beech wood used for compressive characterisation, after having been tested.

cultivation protocol was used across groups: the softening of particles by degradation activities in this context is influenced by the aeration allowed by particles size. The size and shape of particles also influence the matrix shielding. Wood orthotropy and the departure from the linear elastic stress response assumption for the matrix are hypothesised to have a marginal effect on the system given that $E_p >> E_m$ presently (1.19×10^4 MPa $>> 5.5 \times 10^{-1}$ MPa). The matrix discontinuity and heterogeneity are reflected in the tensile elasticity of a matrix considered continuous, and is hypothesised to be primarily influenced by aggregate shape and size for a fixed substrate chemical composition.

The densities for the industrial grade beech wood particles used in the previous experiments are presented in table 3.15. The volume fraction of the particulate types is given for a European beech wood density of 640 kg/m^3 at 12 % MC (D. W. Green *et al.* 1999). Across particle types, the volume fraction is in the 0.281 – 0.516 range and the BM class shows the highest volume fraction. The ratio of mean dried sample density to the beech wood particles density is indicative of the maximal amount of wood substrate having been used in the manufacturing. As the *Ganoderma pfeifferi* decay of the particles and weight loss over a 16 weeks fermentation period (four times the one used in this work) at 22 °C on solid European beech wood blocks has been reported to be of 13.4 % (Szczepkowski and Konsencjusz 2006), the particles volume loss in our experimental works is considered marginal. The maximal volume fraction ratio is represented by the relation:

$$\varphi_p = \frac{\rho_p}{\rho_w} \times \frac{\rho_c}{\rho_p} = \frac{\rho_c}{\rho_w} \qquad (3.5)$$

With φ_p as the particles volume fraction in the composite, ρ_p the bulk density of the particles, ρ_w the density of the wood and ρ_c the density of the composite. The maximum particle volume fraction is determined to be in 0.289 – 0.365 across homogeneous specimen groups (table 3.16).

From a study reported in the literature, the substrate volume ratio of corn stover fibres in a mycoboscus was determined to be in 0.28 – 0.31 (Islam *et al.* 2018a). The elastic modulus of the matrix can be expressed from the previous equation for the inverse rule of mixture:

$$E_m \geq E_{c-} \times \varphi_m \times (1 - \frac{E_{c-} \times \varphi_p}{E_p})^{-1} \tag{3.6}$$

A Young's modulus for European beech wood of E_p = 11.9 GPa at 12 % MC (D. W. Green *et al.* 1999), and $\varphi_m = 1 - \varphi_p$ were used. In table 3.16, the maximum particle volume fraction is indicated for each specimen group. The matrix modulus is approximated and is then normalised by the mean group density to approach the specific matrix modulus. The minimal determined values for E_m are 0.39 MPa (BS) and 0.55 MPa (BM), which reflect matrix averages. The compressive specific modulus is similar between the BM and BL groups, which challenges the conclusion that the BM particle size is the only best performing. The lower compressive specific modulus for the BSML group supports the hypothesis that small-sized particles partially prevent aeration during fermentation which might lead to a less dense mycelium. The tensile specific matrix modulus is of a comparable order between BS and BM groups, to the advantage of BM. The difference between the approached matrix moduli and the pure mycelium values reported in table 3.16 can be explained by both the approximations in the use of the rule of mixture and the very different cultivation conditions between the two experimental setups. It was also previously noted that important phenotypic differences can be found between different strains of a fungal species (section 2.4). Lastly, as E_m values of 0.6 – 2.0 MPa were determined for mycelium densities of 30 – 50 kg/m^3 with an evolution of E_m with an undisclosed square relationship (Islam *et al.* 2017), it can be hypothesised that the mycelium grown density in the BM specimens was < 30 kg/m^3.

Figure 3.43: Section of a mycoboscus cultivated on medium sized particles.

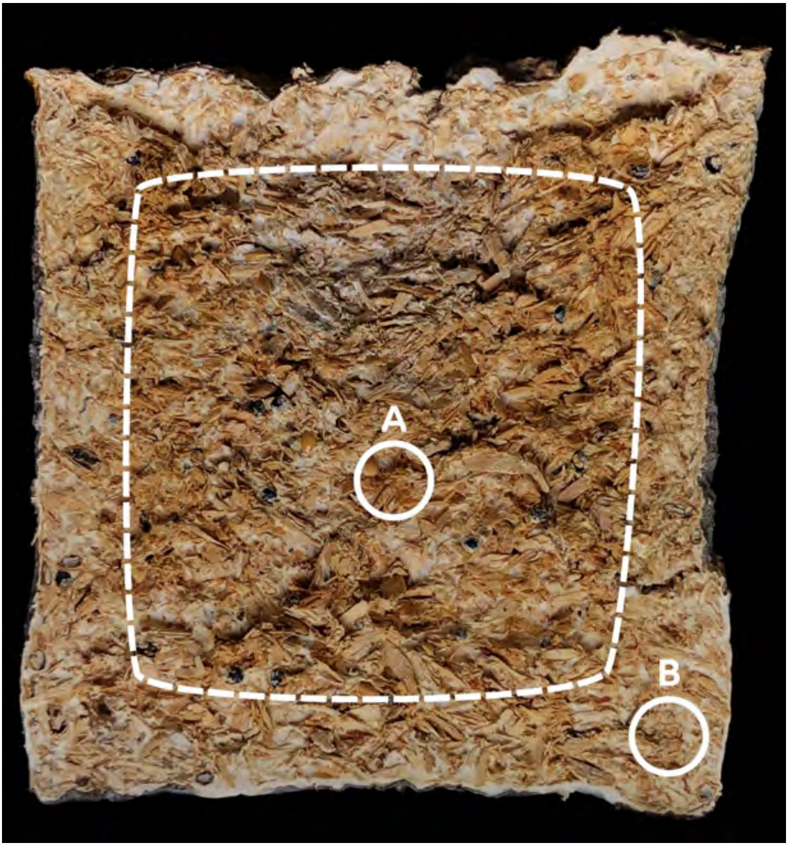

Figure 3.44: Section of a mycoboscus cultivated for four months on spent substrates. The area within the dashed curve is typically less fermented and colonised with a less dense mycelium: in the area A the particles contain more lignin and look more brown, while in area B a very dense mycelium can be observed and the particles are whiter.

Type	**Granulate size**	**Density** (kg/m^3)	**Volume fraction**
BS	0.5 – 1.0 mm	180 – 250	0.281 – 0.391
BM	0.75 – 3.0 mm	270 – 330	0.422 – 0.516
BL	4.0 – 12.0 mm	220 – 320	0.344 – 0.500

Table 3.15: Nominal densities given by the supplier of the European beech wood particles, and nominal volume fraction given for a European beech wood density of 640 kg/m^3 at 12 % MC (D. W. Green *et al.* 1999).

Stress case	**Particle type**	**Mean density** (kg/m^3)	**Mean Young's modulus** (MPa)	**Maximum particles volume fraction**	$\mathbf{E_m}$ (MPa)	**Specific matrix modulus** ($10^3 m^2 s^{-2}$)
Compression	BS	209.67	1.79	0.328	1.20	0.0057
	BM	233.87	3.32	0.365	2.10	0.0090
	BL	217.60	2.96	0.340	1.95	0.0090
	BSML	220.59	2.17	0.345	1.42	0.0064
Flexion	BM	216.27	192.71	0.338	127.57	0.5899
Tension	BS	185.15	0.55	0.289	0.39	0.0021
	BM	217.35	0.84	0.340	0.55	0.0025

Table 3.16: Summary of specimen types parameters, resulting dried densities, elastic modulus, maximum particle volume fraction, approached matrix elasticity and specific elasticity.

3.5 Conclusion

From the FTIR substrate analysis of the strain *G. lucidum* used in the study, it was shown that there is a preference in the break down of hemicellulose and lignin. This is coherent with the state-of-the-art and in line with its known life-history strategy as a selective decay species.

Both in compression and tension the performance of particles of the smallest granulation (BS) was significantly lower than that of other granulations with regards to ultimate strength and elastic modulus (α=0.05). The finer grinding of wood aggregates contributes to softening the individual particles; this is due to particles resulting from the shredding process being more elongated, on average. A picture of BS, BM, and BL particles is presented in Fig.3.45 for comparison. While being consistent industrial products, the granulates display considerable geometrical variations between scales: from small elongated fibres, to flaky planar medium-sized particles, to bulky and angular large granulates. Moreover, the nominal volume fraction of the BM aggregate type is reported to be 1.50 – 1.32 times higher than that of the BS type (table 3.15).

The analytical method presented in section 3.4.5 for the determination of the matrix specific modulus is useful to compare the binding quality of the grown mycelium for isotropic specimen groups. This metric assumes a continuous matrix phase and equal stress distribution in the composite, and it does not consider matrix shielding. The shape and size of aggregates is considered a dominant variable in non-densified mycobosci as it effects matrix shielding (Lu *et al.* 2020) and the aeration of the system, itself effecting both aggregate softening and the grown mycelium density. The compressive specific matrix moduli of the BM and BL groups being equal, it can be concluded that the fermentation and binding are of a similar quality and mechanical performance in relation to elasticity. These groups resulted in mean specific matrix moduli that are 58 % higher than that of the compression BS group and 41 % higher than that of the BSML group. In tension, the BM group specific matrix modulus is 19 % higher than that of the BS group.

Based on the beech wood aggregates datasheet provided by the supplier, the BS class is composed of a maximum of 90 % by particles of a size $\leq$ 0.5 mm, while the BM class is composed at a maximum of 95 % by particles of a size $\leq$ 1.0 mm (appendices B.1 and B.2). With comparable aggregate sizes, Douglas fir wood (*Pseudotsuga menziesii*) particles of a mass-averaged diameter in 193 – 781 μm (mean length of 0.32 – 2.46 mm, mean width of 0.18 – 0.8 mm) were reported to have increasing loose and

Figure 3.45: Small (BS), medium (BM), and large (BL) granulations of beech wood (2021).

tapped densities and decreasing porosity with increasing aggregate size (Tannous *et al.* 2013), although the present normalisation by density nullifies this effect. It was reported that wood aggregate sphericity decreased when mean particle diameter increased (Tannous *et al.* 2013), and the volume fraction of a powder decreases with increasing particle angularity (Guo *et al.* 1985). The flow ability and gas permeability of wood powders increase with the particle size in this dimensional interval (Tannous *et al.* 2013; Geldart 1973). The 29 % lower compressive specific matrix modulus between the BSML and BL groups, and the 12 % increase between the BSML and BS groups contribute to corroborate the hypotheses underlying the material model regarding the presence of smaller particles hindering gas exchange and limiting mycelial development.
This analytical method for the determining of the specific matrix modulus is presented here for the first time regarding mycobosci, to the best of the author's knowledge. This metric can be used to evaluate the quality of the grown mycelium matrix and identify dominant variables based on the system of study, for non-densified composites.

Composition-wise, three typologies were studied in compression: rattan fibres perpendicular to load, common reed fibres coaxial to load, and hessian jacketing coaxial to load. The addition of fibre coaxial to load had a significant effect over the Young's modulus and ultimate strength (α=0.05). Because of the wide range in particle sizes used and fibre composition typologies, the significant difference between specimen groups supports the hypothesis that the two-phase particulate model is suited for future mycobosci studies (α=0.005). From this observation, future studies might involve exploring a wider variety of particle shapes, natures, and distributions as these parameters have been shown to have a significant influence over the elastic and plastic behaviour of composites (Shen *et al.* 1994). The modifying of specimens was demonstrated to be useful for attaining contrasting examples of coaxial reinforcement and perpendicular fracture initiators, with significant effect (α=0.005). However, it should be noted that fibre placements were subjected to variability as fibres could partially misalign with the load axis, or its perpendicular, during specimen manufacturing. This suggests that the standard deviation of the results can be reduced by improving the accuracy in production protocol.

Three fibre composition designs have been reported for flexural stress response and show significant increases in stiffness and strength comparing the BM_R and BM_H series to the BM control group (α=0.05). The BM_I group, with a layer of hessian inserted at mid-thickness of the specimens, did not result in a significant effect. The mean of the flexural modulus for the most performative BM_R series (1.34 GPa) is still close to three times lower than that of Medium-Density Fibreboard (MDF; 4 GPa), and approximately 16 times lower in modulus of rupture (0.62 MPa, MDF: 10 MPa). Considering the higher density of MDF (750 kg/m^3) as compared to the BM_R series (249.48 kg/m^3) a future investigation could focus on developing a hybrid strategy of composition and densification so as to improve aggregate interlocking and resulting strength and stiffness. Composite manufacturing accuracy improvements are expected to contribute to reducing the standard deviation of results. Furthermore, reinforcements may be strategised in developing efficient or multi-functional composites, for instance, in designing the principal substrate to be thermally performative and introducing reinforcements to perform structurally. Such strategies could be used to mitigate manufacturing risks: if a composite is subjected to production inconsistencies, its composition may increase its stiffness and user safety. Moreover, a future investigation could be to hybridise strategies of composition and densification so as to improve aggregate interlocking and resulting strength. The arming strategy adopted with the introduction of inner hessian did not prove significant, as it was positioned at mid-height of the specimens. It was, therefore, theoretically not subjected to tension and could only partially contribute to the bending stiffness. A future iteration of the experiment may look into different soft arming positioning strategies by reflecting on the most mechanically demanded areas of the specimens. The flexural elastic modulus of rattan reinforced specimens, with a specific stiffness of $5.37\ 10^3 m^2 s^{-2}$, along with the hessian jacketed group and its specific stiffness of $1.59\ 10^3 m^2 s^{-2}$, are situated above other reports of the state-of-the-art (Fig.3.48, Fig.3.49). This indicates the wide design space left to explore by isolated or combined use of design strategies. The dimensional stability after drying as linked to diverse reinforcement strategies was presented; this aspect can be investigated in the future both as a means to explore its design consequences, and for production control.

Reports from densification strategies, happening at substrate preparation or by cold or hot-pressing, display a larger increase in ultimate strength than stiffness compared to composition techniques. The addition of reed fibres coaxial to the load case have been shown to increase both stiffness and strength in a balanced manner. The addition of these hollow fibres contributed to a slight decrease in density, thus resulting in a more efficient performance tuning. Substrate supplementation has also been shown to increase performances without influencing density.

Comparing them to the compression results of BM specimens, the change in fungal species from *G. lucidum* to *G. resinaceum* for an identical experimental protocol resulted in a significantly lower strength and elasticity. To tie this result to section 2.8, a comparative study of isolated mycelia from these two species could be predicted to display a greater elasticity of *G. resinaceum* mycelium, thus expanding the manufacturable material properties. As expected from the enzymatic activity review, and although *G. lucidum* is one of the most versatile selective delignifiers, specimens cultivated on pine wood sawdust showed a limited colonisation density and lower performance. Cultivation protocols on gymnospermous woods appear to necessitate changes to those applied on hardwoods, but with the statistical significance of this experiment being non-satisfactory it should be conducted again to be conclusive.

Ashby maps for stiffness and strength as functions of composite density are presented in Fig.3.46 and Fig.3.47. Experimental values reported in this work are plotted along with the values from the state-of-the-art covering the references presented in table 2.1. The distribution of specific stiffness to specific strength is presented in Fig.3.48. The distribution of species and substrates from the state-of-the-art and the present work is presented in Fig.3.49 and Fig.3.50. The same dataset is presented for specific stiffness per species in Fig.3.51, specific strength per species in Fig.3.52, specific stiffness per substrates in Fig.**??**, and specific strength per substrate in Fig.**??**.

Other mycoboscus mechanical characteristics remain under-studied, such as hardness. For instance, a recent study has reported that a *Ganoderma australe*-based mycoboscus could endure a point load of 379.4 N compared to 51.7 N for EPS (Aquino *et al.* 2022). Studying further the fatigue behaviour, weathering and hardness will prove useful for improving market-readiness.
The study report on here made use of Austrian beech wood, hessian manufactured and cultivated in France, and rattan fibres produced in South-East Asia and West Afrika regions (Zhao *et al.* 2017). Beyond the interest of rattan fibres for their very consistent dimensions that help supporting reproducibility of results and material homogeneity, future market-orientated mycobosci developments may focus on constraining the geographical footprint of their attached supply chains so to reduce embedded energy and global environmental stress.

Across the chapters II and III, the principles driving sustainable mycobosci design and engineering were identified and it was attempted to render these comprehensibly. Environmental risks were identified in chapter II and the strategies presented in chapter III are presented as a technical contribution to mitigating these. Beyond objectivised technological design activities, the strategies for the diffusion of the uses of mycobosci may also influence the risks mitigation. This is studied in the following chapter, and takes for a starting point of study the analysis of the effect that an industrialisation process has on this practical aspect. Continuing through review works, the notion of knowledge production, maintenance, and its access to the greater number of people is identified as a central point of interest for the possibility of an eco-social sustainability. Building on this finding, an alternative diffusion strategy as democratic is hatched.

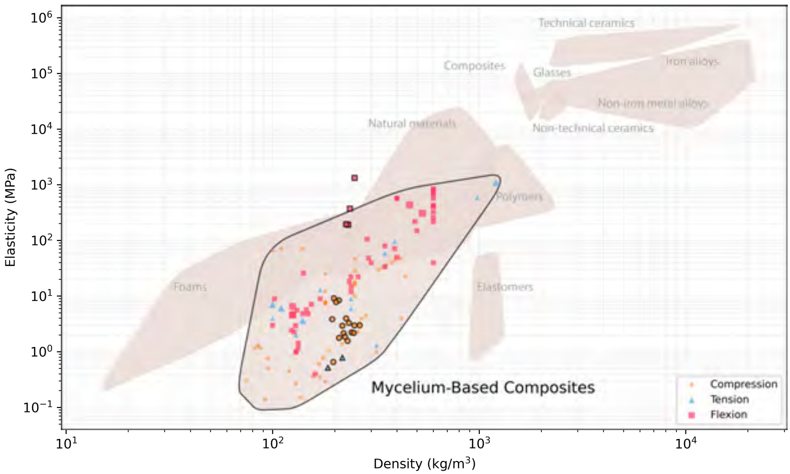

Figure 3.46: Elasticity as a function of density. Contoured points correspond to the compression series reported in section 3.4.1, tension series in section 3.4.2 and the flexion series reported in section 3.4.3. Larger markers indicate composition strategies.

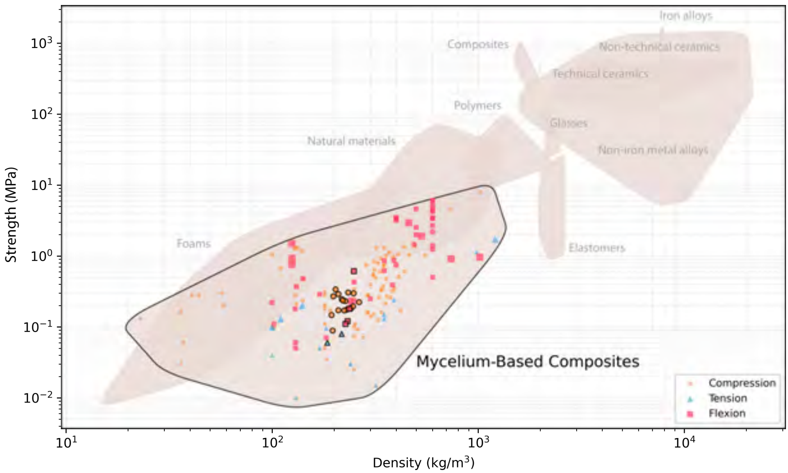

Figure 3.47: Strength as a function of density. Contoured points correspond to the experimental values reported in this study. Larger markers indicate composition strategies.

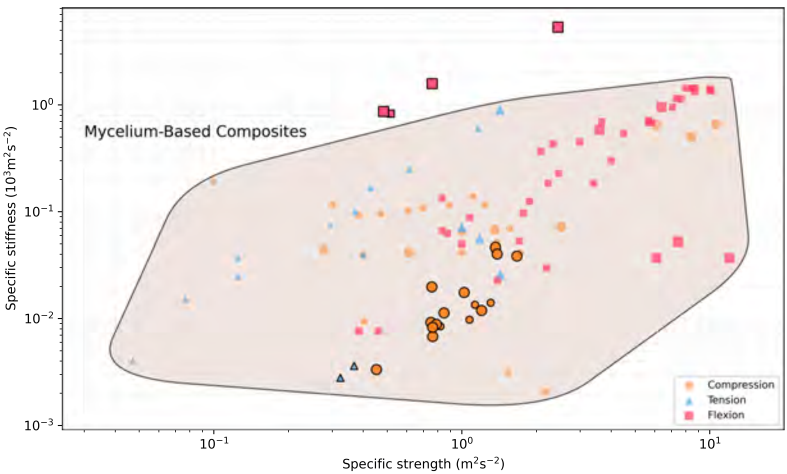

Figure 3.48: Specific stiffness as a function of specific strength. Contoured points correspond to the experimental values reported in this study. Larger markers indicate composition strategies.

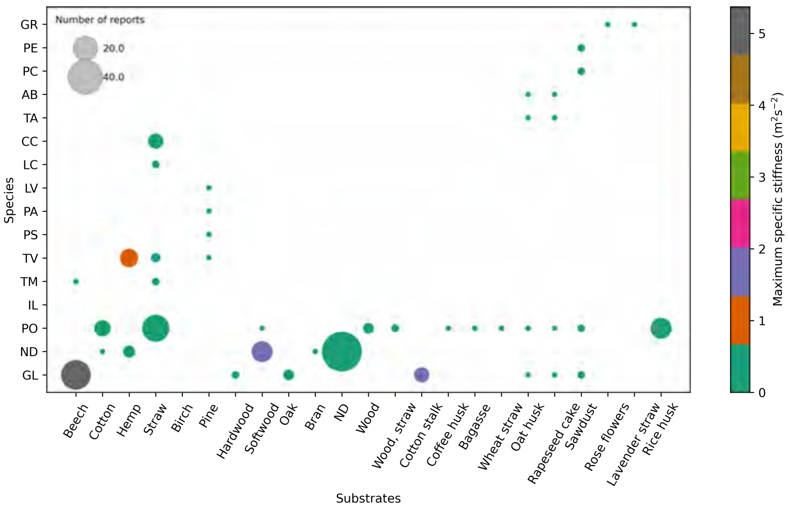

Figure 3.49: Distribution of the mycobosci state-of-the-art including the values reported presently. Larger markers indicate composition strategies. GR: *Ganoderma resinaceum*, PE: *Pleurotus eryngii*, PC: *Pleurotus citrinopileatus*, AB: *Agaricus bisporus*, TA: *Trichoderma asperellum*, CC: *Coprinus comatus*, LC: *Lentinus crinitus*, LV: *Lentinus velutinus*, PA: *Pleurotus albidus*, PS: *Pleurotus sanguineusm*, TV: *Trametes versicolor*, TM: *Trametes multicolor*, IL: *Irpex lacteus*, PO:*Pleurotus ostreatus*, ND: non-disclosed, GL: *Ganoderma lucidum*. The species are listed in Table 2.1 with the reference of the studies.

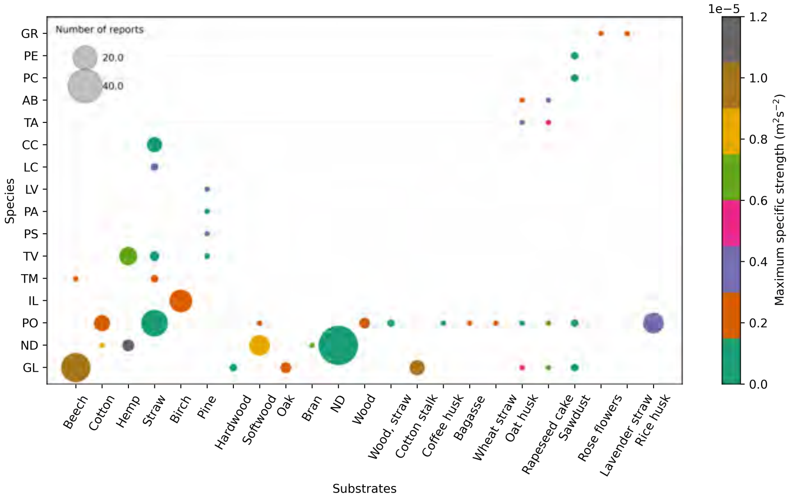

Figure 3.50: Distribution of the mycobosci state-of-the-art including the values reported presently. Larger markers indicate composition strategies. GR: *Ganoderma resinaceum*, PE: *Pleurotus eryngii*, PC: *Pleurotus citrinopileatus*, AB: *Agaricus bisporus*, TA: *Trichoderma asperellum*, CC: *Coprinus comatus*, LC: *Lentinus crinitus*, LV: *Lentinus velutinus*, PA: *Pleurotus albidus*, PS: *Pleurotus sanguineusm*, TV: *Trametes versicolor*, TM: *Trametes multicolor*, IL: *Irpex lacteus*, PO:*Pleurotus ostreatus*, ND: non-disclosed, GL: *Ganoderma lucidum*. The species are listed in Table 2.1 with the reference of the studies.

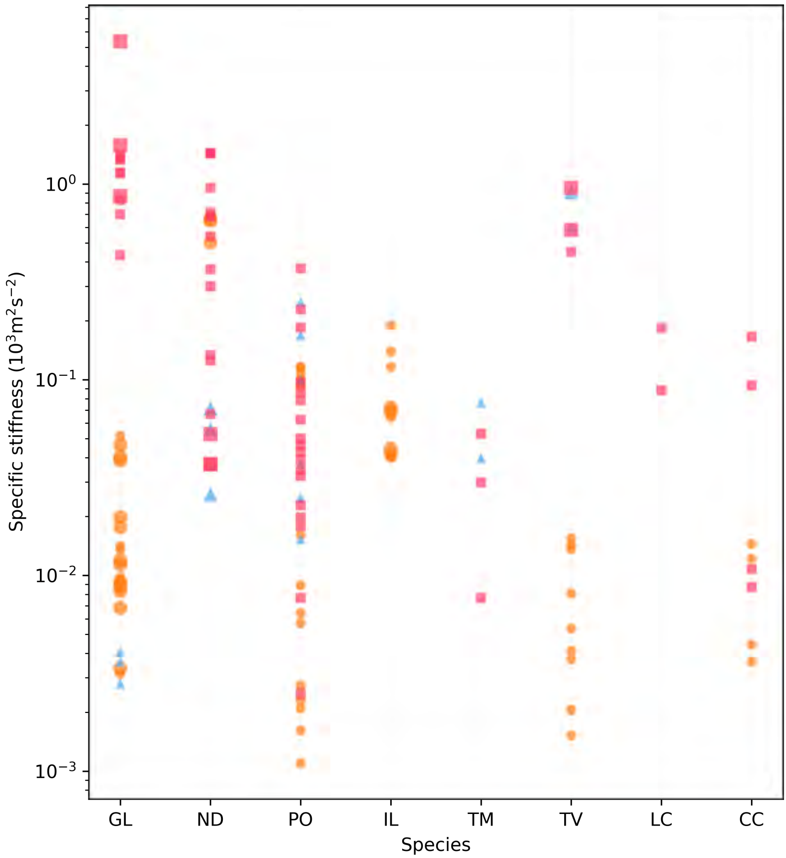

Figure 3.51: Specific stiffness in the mycobosci state-of-the-art including the values reported presently, classified per species. Larger markers indicate composition strategies. GR: *Ganoderma resinaceum*, PE: *Pleurotus eryngii*, PC: *Pleurotus citrinopileatus*, AB: *Agaricus bisporus*, TA: *Trichoderma asperellum*, CC: *Coprinus comatus*, LC: *Lentinus crinitus*, LV: *Lentinus velutinus*, PA: *Pleurotus albidus*, PS: *Pleurotus sanguineusm*, TV: *Trametes versicolor*, TM: *Trametes multicolor*, IL: *Irpex lacteus*, PO:*Pleurotus ostreatus*, ND: non-disclosed, GL: *Ganoderma lucidum*. The species are listed in Table 2.1 with the reference of the studies.

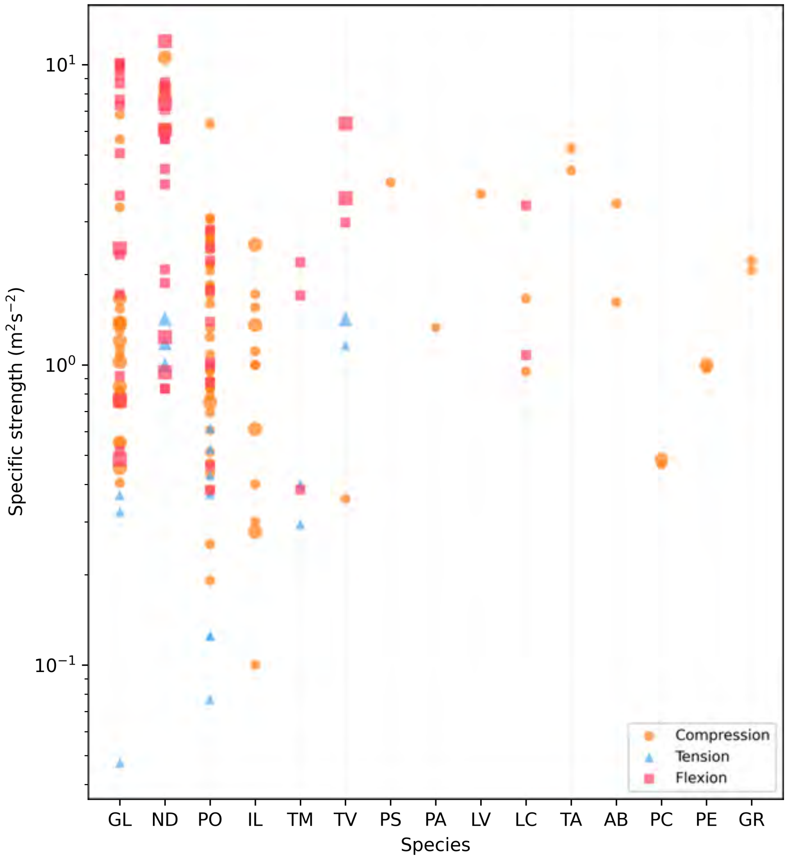

Figure 3.52: Specific strength in the mycobosci state-of-the-art including the values reported presently, classified per species. Larger markers indicate composition strategies. GR: *Ganoderma resinaceum*, PE: *Pleurotus eryngii*, PC: *Pleurotus citrinopileatus*, AB: *Agaricus bisporus*, TA: *Trichoderma asperellum*, CC: *Coprinus comatus*, LC: *Lentinus crinitus*, LV: *Lentinus velutinus*, PA: *Pleurotus albidus*, PS: *Pleurotus sanguineusm*, TV: *Trametes versicolor*, TM: *Trametes multicolor*, IL: *Irpex lacteus*, PO:*Pleurotus ostreatus*, ND: non-disclosed, GL: *Ganoderma lucidum*. The species are listed in Table 2.1 with the reference of the studies.

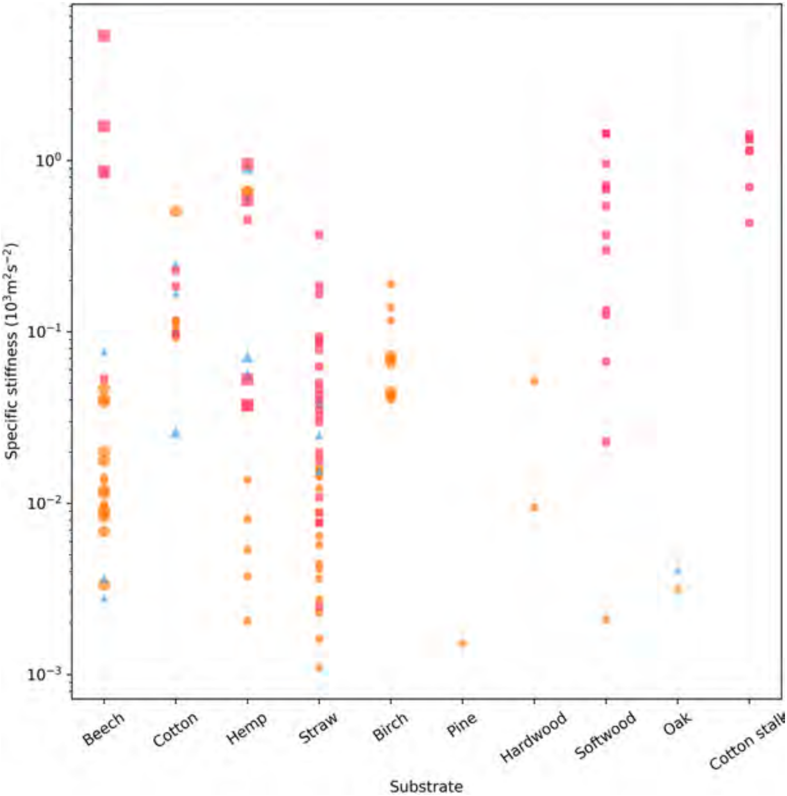

Figure 3.53: Specific stiffness and strength in the mycobosci state-of-the-art including the values reported presently, classified per substrates. Larger markers indicate composition strategies.

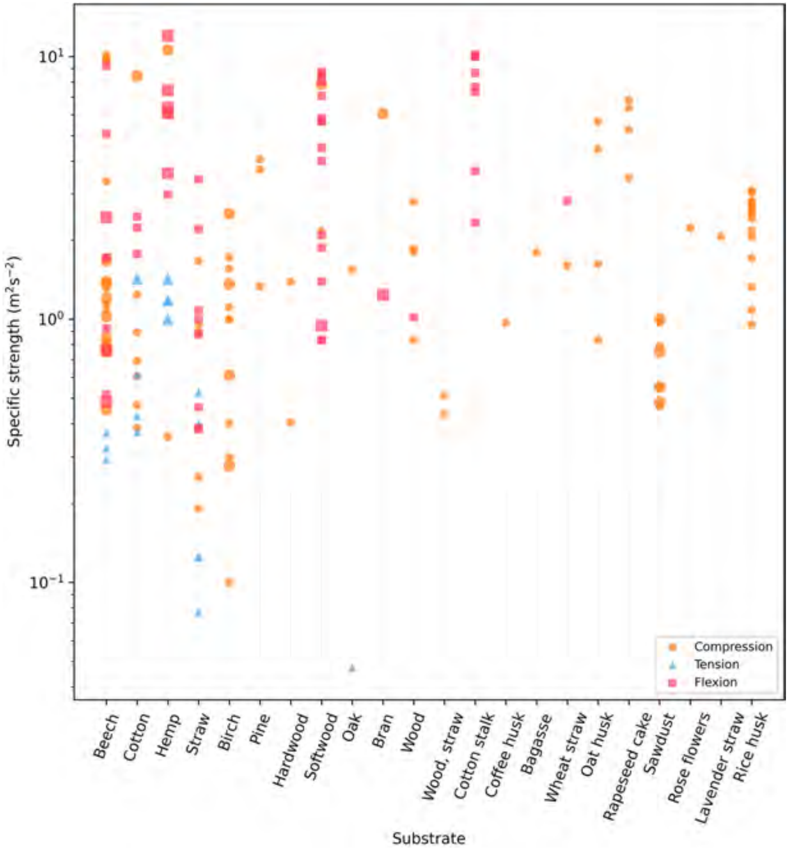

Figure 3.54: Specific stiffness and strength in the mycobosci state-of-the-art including the values reported presently, classified per substrates. Larger markers indicate composition strategies.

Chapter IV – Relational Design

4.1 Introduction

Mycobosci are yet to be fully explored and understood as per their design capabilities. Among the three principal design strategies for mycoboscus that were identified previously (section 2.10), the supplementation track is particularly far from having been completely studied to date. While mycobosci state-of-the-art shows a relative reproducibility to obtain materials with a compressive modulus of c. 4 MPa (median) and compressive strength of c. 300 kPa (median) (table 2.8), making it at least partly technically competitive against expanded or extruded polystyrene for some use-cases, it remains a low-performance material in terms of its mechanical capabilities[30]. These composites are typically envisioned for use in packaging, sound absorption and thermal insulation applications now, to the exception of more creative research (such as illustrated in Ayres *et al.* 2023; Rigobello and Gaudillière-Jami 2021). But it can be otherwise, as the wealth of cultivation parameters that were discussed previously across the design strategies either displayed significant performance increases, or are expected to.

[30] On Fig.3.46 and Fig.3.47 the plotting of the mycobosci state-of-the-art situates generally below the elastomer and natural materials class for strength and elasticity.

As a new material class, its interest for designers extends beyond engineering, to cultural influence. The enzymatic versatility of fungal species make it a particularly remarkable system, for it can be based on opportunistic, vernacular raw materials. Cellulose-based sources such as second-hand cotton textiles can be used in the design of these materials for example, and display a range of performances and aesthetic properties to answer various needs and desires. While the large distribution of performances in the state-of-the-art is a good indication of a lack of consistent use-case attractors and reproducible methodologies (Fig.3.46, Fig.3.47), it is also an argument for the versatility of the system. Applied on texture works for instance, the learnings from the cultivation principles can help to ripen a fine tuned surface of soft mycelium on an artefact boundaries. Primordia initiation can be controlled too and functionalised for example, the illustration of a prototype is presented in Fig.4.55.

The recent and growing interest for mycoboscus fermentation reflects in commercial interests. Entrepreneurs looking for sustainable designs may be enticed to develop business cases, narratives and supply chains leveraging the potential of these materials. While small and medium-sized professional structures may emerge, the industrialisation of mycoboscus solutions has also been developing for about 15 years now. Only a few actors are operating at this scale, or on this market at the moment, which may come as a surprise considering the higher technical affordability of mycobosci. Because of the central role of the industry in the global economy, the effects of the use diffusion by industrialisation and the functioning of the instruments of the latter may impact the actual sustainability of these solutions along with societal adoption. In this chapter, the impact of the industrial development of mycobosci on the mitigation of the environmental risks identified in chapter II is evaluated by way of an integrative literature review (section 1.3). Building on the findings that

Figure 4.55: Detail of a mycelial spirit case with ripened primordia. Own work in collaboration with You-Wen Ji (2022).

suggest an hindering of this possibility, an alternative diffusion strategy that lifts this hindering is investigated with the same integrative method. Hypotheses are drawn from the results of this second part of the chapter, and experimentally tested in a third part.

The first part of this study is interested in identifying the principal effects on sustainability and societal adoption that are inherent to an industrialisation process. To this end, the study focuses on industrial scaling. System scalability borrows from various meanings depending on the field and context, and may hence be implemented with a variety of methods (Putnik *et al.* 2013). The base principle is nonetheless simple: scalability means that the system capacity can be expanded or reduced efficiently in function to its resource use (Heddaya 2002). Once this technical quality is rendered, business intelligence (Negash 2004; Yalcin *et al.* 2022) may be employed to inform an evidence-based marketisation and product lifecycle management (PLM). PLM is a business and industrial management area focused on maximising operational margins leveraging the flexibility, scalability of the value chain (Stark 2022; Terzi *et al.* 2010). These practices illustrate the complexity of the matter in industrial scalability and its permanent dependence to economic growth for profit optimisation. In the context of the diffusion of mycoboscus as thought to be sustainable, this growth relay in the PLM planning conforms to the green growth theory. To comprehensively identify some of the foundational dynamics of the role of scalability in industrial development, a first introduction to green growth is proposed. The study continues with a historical view of industrialisation, with a focus on its epistemology[31]. The PLM practices, among others, also tie intrinsically the industrial capacity to the market to insure a return on investment for capital expenditures (investment costs). PLM practices consisting in maximising the profits for a product on the market, they are a cornerstone activity to commodification (Appadurai

[31] Epistemology is the branch of philosophy interested in knowledge and its production.

1994). By making use of a popular innovation tool for market positioning, the blue ocean strategy, the possibility of sustainable practices is then discussed.

The efficacy of industrial engineering practices reflects in a contemporary global and deep entanglement of infrastructures. These practices are based on scientific objectivisation, and part of their success lies in the generalisation of the inputs of the value chain. Economical and anthropological studies are reviewed to better qualify the state of this entanglement, and the effects of objectivisation on sustainability and public adoption. Along with a quantitative economic study, the philosophical and cultural effects of global industrialisation and commodification of the landscape are then brought forth and analysed. The current industrialisation efforts for mycobosci are then presented in the perspective of the recently established taxonomy for negative commons. This concept is contextualised in relation to the notion of commons and the process of their establishment, where the making of commons is defined as a politicisation of socio-environmental controversies.

The second part investigates an alternative to the industrial diffusion strategy as it proves adversarial to the conditions of sustainable practices development and public adoption. For this, affordable designs living by societal and geographical conditions are reviewed. The scoping of cases used for the analysis was done for their bioregionalist interest. Bioregionalism is a movement having started in the nineteen-seventies at the confluence of a growing interest for biogeography and the development of a counter-culture in California (Alexander 1996). One of the definitions for a bioregion was proposed by Peter Berg, initiator of the movement, and Raymond Dasmann in 1977:

> [It] refers both to a geographical terrain and a terrain of consciousness—to a place and the ideas that have developed about how to live in that place. Within a bioregion the con-

> ditions that influence life are similar and these in turn have influenced human occupancy.
> (Berg and Dasmann 2014 p.218)

Bioregionalism valorises the cultural geography framing and promotes the increased sustainability of politics and economics organised around naturally defined regions. These regions can be defined from environmental landmarks such as watersheds, soil profiles or ecological interactions (Alexander 1996). The rapprochement of geographical and social attention in this movement finds a research interest for mycobosci given the epistemological conclusions from the industrialisation context review, and the greater affordability of their cultivation process. Vernacular architecture cases are then mobilised to identify design characteristics that can support the development of an alternative design diffusion strategies tying environmental management and epistemological justice. As a deeply bioregionalist exemplar, the conditions for the mode of existence of terroir are reviewed and made central to elaborate an eco-social context for the democratisation of the craft of mycoboscus.

The third and last part of this study addresses the epistemological hypotheses established previously. Using the context of architectural education and acknowledging the sense-building focus of this discipline, a pedagogical experiment was conducted to analyse the effects of different epistemological conceptualisation of knowledge conveyors.

4.2 Context

4.2.1 Introduction

While mycobosci can be designed so as to both adhere to circular economy principles and be neutral as per CO_2 emissions equivalence throughout their lifespan, the current dynamics for the diffusion of the technology may not use this capacity. Furthermore, other LCA metrics than CO_2 emissions equivalence should be integrated to avoid impact transfer (section 2.11). If mycobosci are not to become another environmentally risking production activity, the diffusion of their use should consider methods for preventing the previously identified risks tied to their cultivation (section 2.12). In summary, the risks comprise the:

- Global dispersal of chlamydospores of ecologically competitive species,
- Development of plant monocultures for mycobosci substrates,
- Increase in land use potential,
- Increase in eutrophication potential,
- Increase in acidification potential,
- Energy use during cultivation and its polluting effect.

In the context of the current monopolistic industrial development that the company Ecovative unfolds, none of the sustainable development aspects above are considered. The company advertises its use of hemp byproducts as a supposedly sustainable source of substrate (such as in Holt, McIntyre *et al.* 2012). The fact that the product has a commercial or functional value lifts its waste status. Beyond their commercial use in mycobosci, hemp shives are used for many different applications such as biofuels production, insulation or animal litter (Ji *et al.* 2021; Kremensas *et al.* 2017; Arango *et al.* 2023), and still represent an ecological concern if hemp use and culture was to be developed to the detriment of biodiversity and ecological concerns, especially as it requires the use of fertilisers (Zampori *et al.* 2013; van der Werf 2004).

Recommendations for the sustainable cultivation of mycobosci were made in section 2.12, reflecting on the risks it may present. These summarise as the need for developing:

- Supply chains supporting a local cultivation for local distribution,
- The opportunistic use of byproducts to upcycle,
- The use of locally foraged fungal species,
- Passive, natural or animal-powered cultivation processes.

These recommendations are based on biological and environmental sciences, and set out a context that may go against some contemporary *a priori*. After approximately 150 years of accelerated technological commodification since the beginning of the Second Industrial Revolution, the intimate relation between industrial engineering progress, continued economic growth, and Western livelihood improvement and self capacity for determination has led to naturalise a continuity between industrialisation and freedom (Charbonnier 2020). And this naturalisation of industrialisation has been criticised since its beginning in many aspects[32].

How is an industrial development influencing actual sustainability and development of a product? To answer this question considering the current interests in industrialising mycobosci for the diffusion of their use, this examination will focus on the notion of *scalability* to try and identify some issues that might be critical moving forward.

Green growth is a theory integral to the understanding of the context of contemporary industrialisation. Particularly for solutions deemed sustainable, as for mycobosci. After a brief view into this economic theory, two principal aspects of industrial development are reviewed. The

[32]The following references are curated and limited to the corpus otherwise used in this thesis: Dhakal *et al.* 2022; P.-N. Giraud 2012; Gorz 1992; Illitch 1981; Gramsci 1971; Polanyi 1958.

first concerns the positivist[33] paradigm of industrial development. The second aspect relates to the commodification that it fosters, which is identified as a product of marketing activities to which industrialism is dependent—as it needs buyers in volume to acquire its scaled production. The supposed continuity between industrialisation and freedom is then investigated through a comprehensive economic study spanning the 1990 – 2015 period. The significant results of this research demonstrate that capitalist industrialism has unequally benefited high-income countries over the studied period, increasingly and as per embedded labour, energy, land and CO_2 emissions equivalence. And the effects of industrialisation range beyond what can be described by economic instruments. Proposed by anthropologists, the notions of commons and uncommons are reviewed in the following to analyse the effect of the diffusion of the positivist Western *way of knowing*, or epistemology. The study into the commons mirrors then in a recent proposal for the conceptualisation of negative commons (Bonnet *et al.* 2021 p.29): the polluting, ruining commons that are to be dealt with societally now. The review of this landscape finally finds usefulness to elaborate on and specify the likelihood of environmental risks that mycobosci diffusion may present in an industrial context. Additionally to evaluating the possibility of the development of absolutely sustainable practices, a critical interest for epistemology is drawn about the potential for social development with the emerging uses of this new material subclass.

4.2.2 Green growth

Mycobosci-focused commercial companies come down to Ecovative and its licensees (Grow.bio, USA; Grown.bio, NL; Magical Mushroom Com-

[33] Positivism is an empiricist philosophical theory characterised by its postulate that scientific knowledge is the paradigm of valid knowledge. Positivism bases its methods in reason and logic.

pany, UK; Mogu, IT). Ecovative owns a total of 13 out of 26 granted patents worldwide on the technology as of 2019 (Cerimi *et al.* 2019). It is clear that capital securing by patent protection can help companies and industries to sustain a lower production cost. That is by protecting the increase of their industrial capacity, scaling volumes of production, reducing the share of fixed costs per unit sold and increasing operating margins. This promises to increase margins for an actor, but does not automatically reflects on products pricing (section 4.2.4). The protected economic capital is centralised, both in beneficiaries and space; this challenges the prospect for the realisation of the full cultural and sustainable potential of mycobosci. The argument of capital securing holds to attract investors, venture capitalists, and certain academic partners wanting to reduce risks regarding invested capital and its return.

The use of conventional tools for investment, such as patents, to support products and services that are deemed sustainable, affiliates actors to the *green growth* theory. This theory emerged in 2011 with the report *Towards Green Growth* commissioned by the Organisation for Economic Co-operation and Development (OECD). It has since been elaborated upon in multiple contexts by national and international institutions (Ossewaarde and Ossewaarde-Lowtoo 2020; Jacobs 2013), although it lacks a binding definition to be effective (Hickel and Kallis 2020).
The core concept of green growth relies on a continued relevance of the centrality of the power and cultural influence of economic capital, and a supposedly self-regulating free market which would adapt to evolving market demands and externalities (Samuels and Medema 2005; Fleischacker 2002). While the tools for managing and trading in a liberal economy persist, green growth is founded on a principle of decoupling of the economic growth to resource use and climatic events (OECD 2023).
Everything is presented as if the continued growth of Gross Domestic Product per capita (i.e. purchasing power parity, PPP) would be compat-

ible with drastically regulating global resource extraction while satisfying social equity. The massive disparities in public health and environmental legislation, which global commerce is based upon, are source of increasing national inequalities and landscape depletion for all (Dhakal *et al.* 2022; P.-N. Giraud 2012). Furthermore, non-binding international agreements on climate that are supposed to frame green growth development, such as the 2016 Paris Agreement, fall short of their ambition as they are violated by governments themselves (Copland 2020; Grelley 2022). Green growth discourses hardly convince of their good-willingness or ecological efficacy. When the methods and objectives are questioned, the primacy of profits over transitioning to sustainable alternatives even reveals in open conflicts at times (Baron 2022; Correia 2021).

The intentions behind this theory can be illustrated by Kate Raworth's proposal for piloting a *doughnut economy* (Fig.4.56). This decision-aiding tool illustrates the conditions for a minimal and fair development, reminding of the United-Nations Sustainable Development Goals, and articulates some of the LCA metrics into environmental ceilings. The doughnut economy is conceived as the "*safe and just space for humanity*" in this interval, describing a societal model we could theoretically afford (Raworth 2017). Although this model can illustrate the communicated intent of green growth, its theoretical context differs in that it is not based on the predicate that financial tools and objectives should be continued. Furthermore, the doughnut economy framework has been useful to identify the complexity of respecting the planetary boundaries while ensuring social development minima in a context of Western livelihood maintaining. Important research is conducted now in developing metrics tying the environmental ceiling and social flooring boundaries of the model (Ferretto *et al.* 2022).

Green growth is *economic capital* sustainability, and does not bind any of the actors affiliating to it to societal accountability. That is, particu-

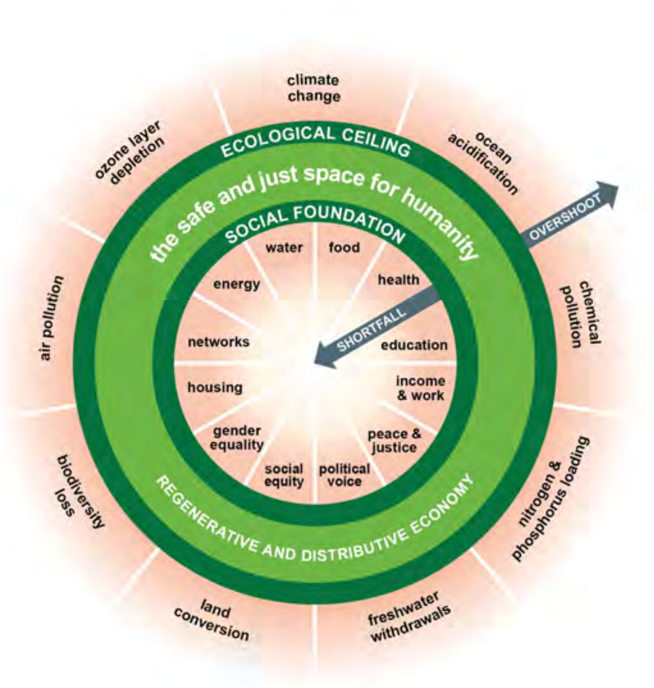

Figure 4.56: Scheme of the doughnut economy. Kate Raworth (2017).

larly in a context where green growth theory is under-defined. Beyond discourses and non-binding promises, the interest in industrial development is that of the scalability of the product or service at hand. The defence of industrialisation for this capacity to be efficient may avoid considering the foundations of the activities it entails. In the following, a brief review of the historical landmarks of industrialisation is made to identify what its effects on sustainability and public adoption are.

4.2.3 Scalability & positivism

In the first half of the XIXth century, in response to the rapid industrial developments of the First Industrial Revolution, Romanticism opposed a holistic and moral scientific culture to deterministic mechanism. Inheriting from the works of Newton on general relativity, Laplacean physics which occupied a central role in the French imperial influence under Napoléon Bonaparte suffered from scientific advances brought by a new generation designated as Fourierists (Tresch 2014). Bringing together an understanding of heat, electromagnetism and optics from substances to processes, Ampère, Fourier and Oersted among other prominent figures, contributed to challenge, through praxis, an opposing of the incompatibility of romanticism to mechanism (Tresch 2014). The design of novel instruments aiming at the scientific description of fluids and energy physics[34] was conceived as a performative parliament of unifying perspectives. Instrument engineering was considered a sensitive and situated practice, strongly influenced by Kantian and post-Kantian philosophy (Tresch 2014; Kant 1998). The often holistic interest of Fourierists is a lasting heritage for opposing a resistive way of knowing to determinism and scientism[35].

[34] As in meteorology, optics, electricity and magnetism, and the description of their relations.

[35] Scientism is the excessive belief in positivism as the best method to describe truth and reality.

In the Second Industrial Revolution, factories started being electrified. The progressive adoption and integration of tool-changing machines that this permitted was concurrent of the advent of a new discipline: methods engineering. Concentrating on the systematic description of mechanical gestures at first, through quantitative explicitation of the physical activity of the chain worker, methods engineers became the functional arm of industrial capitalism. This practice is still very active and has been a keystone of the automatising of manufacturing activities in the XX$^{\text{th}}$ century (David 2011). While industrial machinery have been increasingly integrated and structured by means of technological objectivisation, it reduced supply chains management risks by alleviating a higher entropic factor: human error (Granot 1998). The integration of high-level industrial automation developed with the setting of the Third Industrial Revolution at the dawn of the nineteen-seventies. This was based on programmable logic controllers (PLC) and industrial robotics use, and would later facilitate the flexibilisation (i.e. scalability increase) of global supply chains for financial optimisation. The advent of the lean supply chain model with Toyotism (Dohse *et al.* 1985) and the progression of business intelligence-centred value chains led to the development of an entangled landscape of economic dependencies, coordinated by the use of enterprise resource planning (ERP) software environments (Pollock and Williams 2008).

Based on reproducibility in the concrete or functional forms, industrialisation allowed for the globalisation of the economy. That is by permitting the mobilisation of far situated supply materials to geographically concentrated consumption with emerging demands (Dorninger *et al.* 2021). Energetically and monetarily costly due to resource remoteness and the necessity to maintain increasingly specialised and integrated technologies, the development of mechanical progress and information value chains relied on augmenting volume in market demand by democratising products and services since the end of the XIXth century and throughout the post-war to contemporary period. The success of the industrial model, and a reason for the tendency of cancelling geographic relevance (or cost) in supply chains, can be pre-dated to the gradual adoption of fossil fuels throughout the XIXth century, itself heavily reliant on colonial empires. Being storable, coal, gas and petroleum allowed society to overcome the Malthusian trap—the limits of the organic regime for energy production (Charbonnier 2020 p.129). This contrasts with a previously limited knowledge of the explosive thermo-mechanical power, where water mills, dams and the use of animal power constituted dominating forms of functionnalisable energy sources.

Industrial development has long been the object of critique for its reductionist epistemology. Its scientism has been opposed, at times, to the excess of life that it neglects after the rationalising of a chain worker or a landmark for exerting the patron control (see for instance: Arendt 1958; Polanyi 1958). The nature of this excess is reviewed from an anthropological qualification in section 4.2.6. Contemporarily, the matter of industrial scalability also concerns its dependence to marketing. The blue ocean strategy, a widespread innovation tool, is used to discuss market positioning and commodification dynamics at play under the services that industries make possible.

4.2.4 Scalability & commodification

Innovation management tools can help clarify the underlying strategies of commodification with regards to industrial development, and clear ambiguities that may exist about the use for the quality of scalability. One of these tools, the blue ocean strategy, consists in evaluating and strategising competition-focused product or service market positioning (Kim and Mauborgne 2005). The authors present a simple and articulated taxonomy, with, on the one hand, a *blue ocean* area for innovation, deprived of competition and defended by competitive advantages or higher cost to market access mastered by an actor; and on the other hand, a *red ocean* area representing a market being driven by cost optimisation and pricing competition. An illustration of the blue ocean could be a product that relies on patented technology, for instance, with an exclusive exploitation right granted for a given period. As soon as the technological industrial property falls into the public domain, the product can be exposed to a pricing battle and therefore to cost efficiency that is typical of the red ocean category, if not protected otherwise. A successful establishment of a blue ocean by an actor, or consortium, leads to a monopolistic position by definition, and pricing is based upon their own marketing and research strategies, protecting from potential new coming competitors. For instance, the packaging industry which provides a wealth of product solutions globally, from high end cosmetics, luxury and life science uses, to low cost and larger volume solutions for transporting lower added-value products, is based on a principle of cost control rather than value production. It makes the sector a very economically constrained industry relying on small net margins[36] thus necessitating a critical volume of production based on highly-automated supply chains for mitigating economical risks due to uncertainties in production chain control. The critical volumes of production are managed by economies of scale supported by automated industrial supply chains so that the revenues and operating margins, as reduced as they can become, reach a critical level for at least partly absorbing the production costs. There are two consequences to this: firstly, it becomes obvious in this simplified packaging

[36]The *containers & packaging* industry reported annual net margins of 3.97 – 6.27 % (mean 5.12 %) for the period 2015 – 2019, and returned to 4.53 % in 2021 after a peak at 12.27 % in 2020. In the third quarter of 2022, the trailing twelve months highest net margin was recorded by the *technology* sector at 15.60 %, the lowest appearing in *retail* at 3.00 % (CSIMarket 2022).

example that the scalable quality of the industrial paradigm is defined for a given technical quality of a product; secondly, it suggests that industry and its backing financial systems have long built upon industrial scalability as this is a means and not an end[37]. The first consequence above suggests that by adhering to the green growth theory, one should implement or promote a market positioning for sustainable solutions in a red ocean as this cost competitive area reflects a democratisation of the technology. It comes as no surprise that green growth actors who should be supporting sustainable technologies democratisation are not enacting this, but favour a commodified use diffusion. This can be explained by the need for companies to protect technological knowledge that is critical to maintain a barrier to entrance for potential competitors, and realise their sales previsions. By doing so, they mitigate competition risks for granting a profit return on the invested capital. Different forms of knowledge protection can be adopted for this. The most known legal methods result in patents, copyrights or trademarks, for instance. Other methods have been qualified of strategic, and involve the recourse to secrecy, innovation to market lead time and complex design (de Faria and Sofka 2010). Lead time was found to be generally the most frequent strategy to be implemented, as concluded by a study concerning 1871 multinational companies based in Germany and in Portugal (de Faria and Sofka 2010). This method consists of being first to access the market segment for a solution, which means to ideally open a blue ocean. This method is generally considered to be the most effective for ensuring a market lead with a process innovation (Harabi 1995)—such as with mycoboscus technology. The innovation and industrialisation capacities that support competitive lead time implementation are based on maintaining a difference of technological knowledge with the potential competitors. PLM

[37] More advanced economical strategies have been implemented such as transfer pricing consisting in inter-subsidiaries sales in deficit or at zero net margin to optimise taxes on yearly revenues (Paz and Escobar 2019; Lenz 2020).

is an activity that integrates these dynamics for profits management. It is recognised that actors adhering to the green growth theory generally aim at economic capital sustainability, and not environmental patrimony sustainability (Hickel and Kallis 2020).
Regarding the second consequence of industrial scalability, the main principle of Fordism illustrates it. It consists in paying a workforce a minimum salary so that they can afford the products they contributed to manufacture, and re-inject the capital from their salaries into the acquisition of the product they manufacture (Aglietta 2000 p.116; Dassbach 1991; Adkins 2015; Gramsci 1971 p.311). This shows the advent of a deep and early seeded financial and political entanglement that industry has been naturalising. At the dawn of PLM, a packaging solution is made into a service, a value-based production for the consumer via its optimised supply chain design and maintenance. Reports of operational management in Amazon warehouses clearly illustrate this: the lead time and customer experience subsumes the whole supply chain, rendering it as a slave to Amazon customer satisfaction (Agnihotri and Bhattacharya 2022; Leonhardt 2021). The genius of market positioning has revised the valorisation of the packaging, previously thought to be a relatively independent value chain in itself that was subjected to specific externalities and innovation driving forces, into a packaged instrument of blue ocean positioning expressed and measured in terms of consumer experience and part of a market control strategy. Making a packaging into a selling point for its sustainable quality is probably useful, as it contributes to increase the expectations from consumers with this regard. But if their normalisation is desired by a society then the tentative patenting of underlying technologies should be questioned. This entanglement of industrial scaling with an optimised customer experience is at the core of the maintaining of exploitative systems with regards to social and environmental resources wearing, erosion, ruin. This commodification practice initiated the de-

velopment of design activities from the nineteen-fifties on as a marketing tool, instrumentalising cultural planned obsolescence through trend management (utilising principles of PLM again)[38], although the discipline has since partly matured out of this sole market dependence.

Some arguments can be opposed to this portrait of industry development. The principal ones can be about the global need for automated and economically optimised production methods for both optimising resource use, and sustaining the basic needs of the greater population with more affordable prices. To analyse the effectiveness of industrial capitalism on these two ends, a review of trade metrics is offered in the following section to represent and analyse the state of global exchanges permitted by global industrial supply chains, and better grasp its supporting or hindering of sustainable development.

4.2.5 Unequal exchange

As the result from an inquiry into the impact of global supply chains on the possibility of a sustainable transition, Christian Dorninger and a large European research team have recently published a stark study of natural resources, labour and economic appropriation in global trade. In this research, countries are grouped by income levels, and the distribution of material footprint[39] for these is presented for the year 2015 (Fig.4.57; Dorninger *et al.* 2021). On Fig.4.57 it can be observed that high-income countries have a material footprint that is disproportionate to the rest of the world. The authors continued the study to identify the dynamics supporting this instant picture. They report that high-income countries have largely been sole net importers of both raw material equivalents

[38]The history of the commodification of the "Swiss made" bracelet watch as a fashionable item by Swatch is particularly illustrative (Garel and Mock 2016).

[39]The material footprint of a country is calculated as its final demand in terms of goods and services multiplied by all upstream global material requirements associated with one dollar unit of product (Wiedmann *et al.* 2015).

(over 200 Gt), embodied energy (a high energy footprint corresponding to 226.6 EJ/y) and labour (over 5.5 million p-yeq[40]) (Fig.4.58-a, b, d), as accumulated over the 1990 – 2015 period. Furthermore, these countries represent the largest net trade of embodied land over the period (over 30 billion ha), far ahead of China and India which represent c. 5 billion ha once summed (Fig.4.58-c). The authors point out that China displayed the highest accumulated net trade in Trade in Value Added (TiVA; Wiedmann and Lenzen 2018) during the period (c. 1 900 trillion USD), while being a net provider of embodied materials, energy and labour. Nonetheless, high-income countries represented in 2015 15.5 % of the world population and 64.5 % of the global added-value, while China accounted for 18.7 % of the world population and 12.8 % of the global added-value. Dorninger and the research group conclude that high-income countries benefited from a 12 times average increase in TiVA per embodied labour unit (p-yeq) to the rest of the world between 1990 and 2015. Although environmental controversies are complex in nature and tied to economic expectations negotiated between varied scales (Blaser and de la Cadena 2017), what is clear is that the structure of supply chains exploiting environments that are non-native to high-income countries perpetuate relations of colonial domination (Hickel *et al.* 2022). This power exertion further leads to an accelerating environmental extractivism and ruining, and hinders the possibility of a transition to sustainable environmental management. Anthropologist Alf Hornborg reminds that,

> The naturalization of modern technology assumes many forms and is widespread among people in general. It obscures the obscene global differences in wages and purchasing power that are a condition for the very existence of such technology, granting capitalism's central instruments of exploitation the ideological appearance of political neutrality

[40] p-yeq: person-year equivalents (Simas *et al.* 2015).

> and self-evident virtue.
> (Hornborg 2021)

This contemporary enslaving of the world by high-income countries also shows important metaphysical[41] consequences for indigenous communities. Critical for environmental management and social justice at once, the following section reviews anthropological studies based on field reports for qualifying the effect of scientific objectivism on indigenous epistemologies and ontologies[42].

4.2.6 Globalisation and epistemological justice

Although irreducible to explicit notions (Bookchin 2007; Gorz 1992; Merchant 1980), ecological milieus have been thought as the feeding reservoir of Western societies under the influence of scientific objectivisation, alienating the profound otherness and entanglement of biomes under the rationalist idea of *resource*. Approaching ecological management as a scientific effort to extract and transform products of the landscape, or to try and fix the effects for the Anthropocene, is to engender instruments of investigation and action driven by an idea of scientific progress that is reductionist towards ecological interactions and local communities situated epistemologies and ontologies (Blaser and de la Cadena 2017). As Carolyn Merchant analysed in *The Death of Nature*, the increasing scientisation of the relationships to local and global environments has led to considering these as complexes that appointed experts can ultimately fully comprehend, describe and manage with supposedly negligible ripple effects, and objectivise as systems to *maintain*, *shepherd* or *care for* rationally.

[41] Metaphysics is the branch of philosophy studying the fundamental nature of reality, including the first principles of being or existence, identity, space and time, cause and effect, necessity, and possibility.

[42] Ontology is the study of being, becoming and reality in metaphysics.

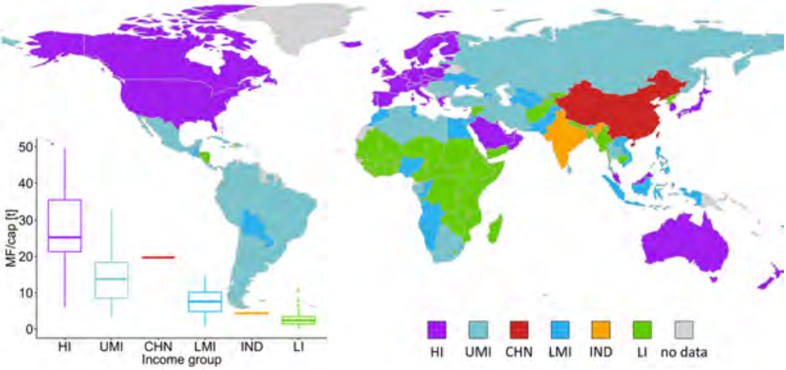

Figure 4.57: The global distribution of income clusters in a map and a boxplot of the distribution of the material footprint in each income-group in 2015. Income-based country grouping: high-income HI, upper-middle income UMI, lower-middle income LMI, low-income LI; China CHN, India IND. Reprinted from Dorninger *et al.* 2021.

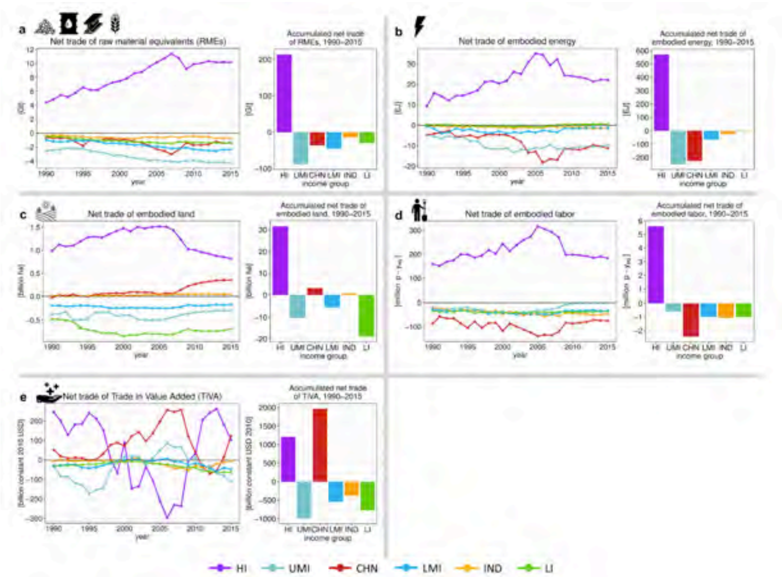

Figure 4.58: Net trade of resources over time and accumulated appropriation and supply as bar plots, 1990 – 2015. a: raw material equivalents (RMEs) [Gt]; b: embodied energy [EJ]; c: embodied land [billion ha]; d: embodied labour [million p-yeq]; and e: trade in value added (TiVA) [billion constant 2010 USD]. Positive values represent a net appropriation of resources. Reprinted from Dorninger *et al.* 2021.

Beyond a critic of scientisation itself, a review of the linear model of expertise adopted by the Intergovernmental Panel on Climate Change (IPCC) was conducted by sociologist Silke Beck to identify potential causes to caveats in its political efficacy (Beck 2011). The linear model of expertise is defined as the unidirectional influence of science over policy making in this study. Dating from 2011, the analysis points to the reluctance of the IPCC towards addressing political implications of scientific consensus, and the lasting panel position to date to favour value-neutral science, which results in proxy debates:

> As numerous negotiation processes inside the IPCC—e.g. concerning the 'value of life'—demonstrate, invoking a univocal and supposedly value-neutral science to resolve such deep-seated normative divergences is a recipe for discord rather than harmony (…). In the end, science is reduced to a spectacle of assessments competing against one another for supremacy—a kind of 'contact sport' (…).
> (Beck 2011)

One recommendation that was made in conclusion, is for the IPCC as experts[43]: "*to provide a robust consensus allowing for continued scrutiny, re-examination, and revision*", for a more participatory, distributed, localised and effective approach to policy informing (Beck 2011). One of the flaws of the linear model of expertise was identified by Beck to be the detachment of expertise from political and cultural contexts, and the separation from "*systems of experience and understanding*". These systems identified by Beck can be further qualified to identify what is neglected.

Under the contemporary influence of cybernetics, ecology is thought as an objectivised activity aiming at managing rationally environments

[43] Expertise is understood in this chapter as the situated, socially legitimated truth production capacity of an individual or group of.

by means of engineering (Hui 2020). The globalising and encompassing nature of cybernetic thinking can be compared to an epistemological colonising effort, cancelling epistemologies differing from its own legacy.

> The social production of nonexistence signals the effacement of entire worlds through a set of epistemological operations concerning knowledge, time, productivity, and ways of thinking about scale and difference.
> (Escobar 2018 p.68)

Here, anthropologist Arturo Escobar brings forth the consequences of these globalising efforts, negating site specifics to the benefit of the Western ontology and power exertion. This leads to the epistemological depriving of engulfed worlds and societies, their shared and inherited ways of knowing, and worldviews. The intimate and foundational conditions of being to the world of situated communities are negated in the rationalist perspective, while their landscape are mined and financiarised in the form of resources and serviceable labour (Dorninger *et al.* 2021).
Investigating various contexts that see these intimate worldviews face off colonisation in various forms, Mario Blaser and Marisol de la Cadena formulated the concept of *uncommons* (Blaser and de la Cadena 2017). The authors describe this concept as "*that which exceeds [the commons] and cannot be articulated by it*", therefore in relation to *commons*[44].
A commons as a mountain may bear a spiritual value to a community, and be a site for intimate stories for indigenous inhabitants—relationally constitutive of the identity of *that* landscape. Once the landmark is integrated into the domain of another network of interest, such as that of an extractive company or a state willing to act *for the common good*, may see its status change to a negative commons (section 4.2.7). Now, the

[44]Commons are cultural and natural resources for which all members of a society are negotiated an access. The commons are traditionally referring to resources deemed critical for the decent living conditions.

objectivised ruins the socio-environmental uncommons by situating the mountain as a mere standing stock of financiarised value to high-income countries.

For formulating the uncommons, the authors build a focus on the negotiation process inherent to the commons, referred to as *commoning* (Bollier and Helfrich 2014). Silvia Federici puts it simply: "*if commoning has any meaning, it must be the production of ourselves as a common subject*" (Federici 2012). Commoning is to politicise and negotiate both what is deemed a resource, and one's conditions of political existence at once. This ties the social becoming—becoming a common, political subject—to cosmological[45] ecological situating, as a continuous process of difference fostering cooperation and responsibility to the other (Federici 2012). The necessity for the recognition of uncommons emerges as the commoning activity may enact in asymmetries of power between stakeholders about a landmark:

> For example, a river like the Mekong might be domained as a common source of livelihood by riverine communities, as a (potential) common source of hydroelectric energy by the governments putatively representing citizens of a country it

[45] In anthropology, cosmology has been defined as being: "*the world view and belief system of a community based upon their understanding of order in the universe*" (Darvill 2008). The notion has become criticised for its aim at cross-cultural scientific holism, consequently conveying the idea of a hierarchy of cosmologies, as modern scientific methods reflect nothing less than an anthropological cosmology in themselves:

> The master contrasts of classical anthropology (tradition/modernity and so on) lend analytical weight to this basic—and for most of the twentieth century self-evident—hierarchy of perspectives. Indeed, the contrast between indigenous cosmology (as something others have), and science (including of course the cosmological research of modern astrophysics) as 'our' ultimately *truer* equivalent, was just another iteration of these master contrasts. (emphasis in the original text; Abramson and Holbraad 2014)

As discussed by Abramson and Holbraad, this notion finds a renewed interest under the current post-structuralist period, by acknowledging that cosmology interests need to beware of holistic essentialism and the conscious or unconscious use of hierarchical categories.

> traverses, and as a key component of the biodiversity commons by transnational environmental organisations. If we keep in sight that the river is a source of livelihood, a potential energy source, or a repository of biodiversity, we can see that each constitutes different projects of domaining (or commoning), and it is easier to grasp the idea that their scales might be an effect of how such projects are pursued.
> (Blaser and de la Cadena 2017)

Casper Bruun Jensen, author of the referenced field study on the Mekong river, further elaborates in the study on the conception of uncommonalities as the essence of the landscape emerging within the domains of commons (Jensen 2017). Jensen suggests that an attempt at negotiating uses through commoning cannot be reduced to the physical environmental actant as the smallest denominator, but to the study of how the domains relate to and influence each other—hence the advent of a politicised subject. In overlooking the uncommons, the intimate and community cosmologies are negated and forcefully replaced, often by Western positivism, a lineage particularly represented by the modernist and cybernetic epistemologies. The environmental object is both materially and cosmologically depleted by way of an asymmetric negotiation between stakeholders[46], and over the longer term the relocation of the colonised within a coloniser cosmology produces an ontological entanglement (Blaser and de la Cadena 2017).

For epistemological justice, Arturo Escobar emphasises on the critical necessity to encourage ontological assemblages and negotiations by commoning and controlled equivocation[47] (Blaser and de la Cadena 2017; Viveiros de Castro 2004). The study of this prescriptive path forward was

[46] In the Mekong river example, governments, riverine communities or transnational environmental organisations do not hold the same power form and amplitude during the negotiations.

[47] Eduardo Viveiros de Castro describes an equivocation as both a failure in understand-

initiated by Blaser and de la Cadena as an investigation into *political ontologies* (Escobar 2018 p.67).
Escobar illustrates the stakes by quoting Francia Márquez, a leader of a march against illegal gold mining in La Toma (Columbia), who wrote in a letter to the government and citizens:

> Everything we have lived, (...) has been for the love for our territories, the love we feel when we see plantain germinate, when we have a sunny fishing day, of knowing our family is close by....Our land is the place where we dream of our future with dignity. Perhaps that's why they [armed actors, including the army, paramilitaries, and guerillas] persecute us, because we want a life of autonomy and not of dependency. (Escobar 2018 p.72)

Márquez' letter insists on the need for epistemological and ontological autonomy, where situated communities have the capacity for auto-determination and their histories respected. Extractivism and the maintaining of international supply chains has been shown to economically and materially benefit high-income countries, degrade the environments and dramatically reduce biodiversity (Dhakal *et al.* 2022). It extracts by exerting an asymmetrical power relationship in commoning, and proceeds in the same arc to sledgehammer the uncommons, the legitimate intimacy of indigenous communities. Sturdy as a leviathan (Weiss 2018), industrial entanglements can be challenged with a turn to a subjectiv-

ing, and a failure in understanding that "*understandings are necessarily not the same*" (Viveiros de Castro 2004). Borrowing from Amerindian perspectivist anthropology, Viveiros de Castro describes an equivocation as foundational to the continuous forming of a relation that it implicates between two actors, forever others to one another. "In this model of translation, (...) difference is therefore a condition of signification and not a hindrance" (Viveiros de Castro 2004). An equivocation relates to Simondon's transduction (Stiegler 1998), it is a relational sense building in difference.

ising[48] relational design—a practice that valorises uncommons by offering a politicised interactionist understanding of design and technology. This turn can extend to the identification of global supply chains as *societally ruining infrastructures*, and open prospects for healing from this. This powerful notion of *ruin*, constructed in line with the understanding of commons and their advent, offers a perspective about citizen agency about the detrimental infrastructures and institutions that are to be addressed. Recently developed within the field of Science and Technology Studies (STS), this notion is outlined in the following section.

4.2.7 Negative commons

Turning to a social understanding of technology and design influences the view on their role in the society, and the idea of progress. The very methods of material and knowledge production involved in the design activities are modified, and may not inherit from previously existing infrastructures and epistemologies.

Conceptualised as a subset of the commons, *negative commons* have been recently introduced in the discussions on post-Anthropocene design. The philosopher of science Alexandre Monnin defines these in the book *Héritage et Fermeture*, as the negative material or immaterial resources, such as wastes, nuclear plants, polluted soils, or certain cultural heritages (Bonnet *et al.* 2021 p.29). The author further elaborates on this idea by proposing the notions of *ruining ruin* and *ruined ruin* (Bonnet *et al.* 2021 p.23). Negative commons qualify as ruining or ruined ruins as an inheritance of extractivist and alienating complexes such as mining supply chains, coal power plants, marginalising health policies or processes of systemic exclusion of habitus deemed unethical for instance.

[48] Subjectivisation is a process in which a design acquires meanings that convey the community or designer's attitude or viewpoint. It is the "*formation of a one that is not a self but is the relation of a self to an other*" (Rancière 1992).

With a foot in social justice and another in environmental justice, each being closely related, the authors of the book argue for the politicisation of previously objectivised industrial facilities and institutions. Furthermore, if associated to environmentally detrimental infrastructures or institutions, emerging solutions presented as *objectively good* may as well present brand new ruins (*ruines flambant neuves*, Bonnet *et al.* 2021 p.35). Solutions and companies consciously or unconsciously adhering to green growth theory for instance may rely on technologies deemed sustainable such as mycobosci, but integrate it in a supply chain and industrial property scheme that fosters capital harnessing, and shadows, or objectivises potential ripple effects. These, would be *brand new ruining ruins* to be dealt with societally. It can further be inferred that negative commons exist alongside *negative uncommons*, or toxic cosmologies—consumption habits, belief systems, epistemologies, philosophies—developing from and around foul commons, or originating them (understood as part of *negative commons* in Bonnet *et al.* 2021).

Living with otherwise, *living with from now on*, and *living without*; these are strategies that compose the scope of methods to address the inherited industrial ruins (Bonnet *et al.* 2021 p.41). While the *living with from now on* scenarios may be fittest to acknowledge scientifically some particular inherited negative commons, such as the anthropogenic increase of droughts and season fires, the *living with otherwise* and *living without* strategies necessitate an engineering of monitored technological transition periods—such as replacing the packaging of all objects sold online by sustainable ones (mycoboscus-based, for example)—or the democratic closing of ruining activities—cancelling online product sales.

4.2.8 Conclusion

As an emerging material class and culture, mycobosci are best democratised with caution as per their technical, logistical and cosmological capacity. The on-going efforts of industrialising these with a patent strategy to avoid the democratic appropriation of the craft reflect a centralised and monopolistic endeavour. Doing so subjects mycobosci to the same positivist paradigm that supports material extraction and use, cosmological colonisation, and which based industrial capitalism and its depleting effects. The risk of spreading active chlamydospores globally, or that of fertilised plant monoculture, and the increase of eutrophication, acidification and land use potentials, seem disregarded by Ecovative given the standardised design of their products[49] and despite their public discourse (Ecovative 2022). These claims of sustainability are greatly influenced by the contemporary green growth theory, both because it is adopted by leading governmental organisations to base their *green deals* (Bloomfield and Steward 2020) thus hiding behind state legitimacy, and is a great niche to industrial actors willing to make promises that are complex to verify by most citizens, given the complicated nature of the technological and economic mode of existence of industrial infrastructures.

About the environmental urgency, *rapid and scalable* solutions are often sought after. But scalability needs redefining from its industrial anchorage and positivist epistemology. Scientific reductionism and its disregarding of local environment specifics, integral to the functioning of a globalised industrial infrastructure, is that which needs evolving from. While the Western positivist tradition has demonstrated the power of its model with a hegemony of high-income countries over global resources extraction (Dhakal *et al.* 2022; Dorninger *et al.* 2021; Hickel *et al.* 2022), unbothered commodification has been a catalyst. If a centralised and

[49]The Grow-It Yourself kit sold globally by Ecovative is using the presence of active chlamydospores within to ship dry and lightweight inoculated substrates, for example.

delegated industrial production model is to be rendered obsolete, at least for mycobosci, the social individuation[50] value of its alternative needs to be considered. For this, designers will have an important role to play as they can add a cultural interest to technology.
In parallel to this, the politicisation of ruining infrastructures and institutions is to be undergone (Bonnet *et al.* 2021; Hornborg 2021; Escobar 2018). This is to question those that may be naturalised, and decide for the closure of ruining ones for technological or social alternatives. The industrialisation process and its impact on the naturalisation of commodified technologies displays a strong power of epistemological and ontological influence, along with a centralised economic power. As such, industrialisation systematically hinders the possibility of a political process about its legitimacy, and represents considerable and unequal material footprints once considered globally. Industrialisation is a ruining human activity. The risks that were identified about mycobosci scaling in volume illustrate how a solution that can be sustainable may result in detrimental effects for the environment, thus making it a *brand new ruining ruin* in potential.
The review of the interest for the establishment and recognising of the uncommons for fair and just living conditions also shows how sensible the political commoning process is to scales and asymmetries of power. As recently discussed by Escobar and supported by Hornborg's studies, stirring the attention of designers to epistemological colonisation by the

[50] The idea of the individuation process has been initially proposed by Gilbert Simondon to describe the process of technical innovation and its integration at the dawn of the advent of integrated tool-changing industrial machines in the nineteen-fifties. This has been elaborated upon both by Simondon and Stiegler in its psychic and social dimensions. This concept of individuation is particularly useful for design studies as it focuses on the effects of the interactions with a technical object on an individual and social groups. The study of individuation is particularly interested in the continuous process of the formation of identity (Simondon 1958; Stiegler 1998). Social individuation, in this context, is the progressive and never completed process of identity forming in one as socially anchored—that is, within a situated community.

industrialisation process is a matter of social justice. Investigating an alternative epistemology of technology is then to try and valorise another mode of existence for designs, where the heritage of Western positivism is politely declined in an effort to live without.

Pursuing a globalised industrial development, furthermore as monopolistic, cannot be a relevant and socio-environmentally sustainable path forward. This, is what needs to change. Its capital-oriented performance assessment, distanciation and reductionism to ecologies, and privatisation of knowledge to benefit capital centralisation, are distinct traits that should rise red flags for entrepreneurs that are concerned with the equitable livability of the planet. What alternative strategies can we devise then, for the craft of mycoboscus to realise its potential of environmental sustainability and contribution to an equitable social development?
The democratisation of their use should start by challenging a privatisation of knowledge and leverage it to support a movement away from commodification and towards local community agency. This adds to the requirements for local cultivation and distribution networks, opportunistic use of byproducts, locally foraged species, and alternative cultivation systems limiting the use of energy. These recommendations make it clear that the local development of cultivation and use networks is important for sustainability and citizens autonomy. Furthermore, the negotiations necessary to the political process of commoning, about the quantitative and qualitative affectation of byproducts to the cultivation of mycobosci for instance, are to be conducted within the scope of bioregions. While the current flows of byproducts concern oil-based single use packaging too, i.e. material flows reaching far beyond the scope of a bioregion, it would be recommendable that this is remediated in favour of compostable materials conforming to biological and geographical contexts.

Subjectivising and situating technologies, as an effort to develop social individuation, will be essential to develop ones that are sensible to

biome specifics (Hui 2020) and that leverage the capacity of communities for auto-determination. After this survey of the qualities of industrial development, the study progresses into designerly contexts to devise an alternative strategy for mycoboscus craft democratisation with a view on bioregions.
Remarkably local and rich of uncommons, the modes of existence of vernacular architecture and terroir are of particular interest for their eco-social designs. What may look to be a remote object of study in terroir—as this quality usually refers to edibles—offers a systemic view of eco-social development efforts, and has been used previously for international development by the United Nations (UN). A synthesis of this effort is proposed in the following section, and it reveals that it was mainly focused on community institution building to facilitate fair commoning and co-becoming with the ecosystems. Fragility, or impermanence, appears to be a structural quality supporting a social individuation in vernacular architecture and terroir. Some forms of frailties are brought forth in the following, and identified as opportunities for commoning and the development of uncommons as cosmologies. Vernacular architecture and terroir are two design areas that are also distinct because of their reference to an alternative authorship model as based on tradition. This offers the opportunity to scrutinise the potential for the development of an alternative epistemology, sensible to individuation as a continuous identity construction.

4.3 Design terroir

4.3.1 Introduction

The focus built on locality at this point reflects in a number of pre-industrial, pre-colonial technological infrastructures. With a reduced technological integration and a restrain from using fossil fuels, the geography and societal organisation have a strong influence on the capacity of the technology to operate. Instead of considering the landscape as a standing resource awaiting extraction and financiarisation, some premodern technologies valorise and depend on the climate, topography, microbial fauna, and social activities to exist. This dependence illustrates a technological fragility, in that the functional range of the technology is reduced, to when sun shines for example. The presence of this may give rise to the organisation of a ritualised maintenance to palliate the fragility, as seen for the Great Mosque of Djenné (Mali) made of rapidly eroding earth plastering (Brunet-Jailly and Scherrer 2018).

It seems unfortunately dualist to oppose point-by-point the mode of existence of the cases discussed in this section to industrialism. But as it will be examined, the qualities of a democratisation strategy that are sought after to try and avoid the structural issues with industrialisation, are indeed present in these bioregionalist designs. Terroir is first described as per its mode of existence and geographical ties, and is shown to reflect both practically and aesthetically[51] in an eco-social relationality. Adding to the logistical interest of the terroir quality articulating the notion of bioregion, an account of its use by the UN helps describing how its implementation has been and can be systematised. Extending the context to vernacular and traditional architecture with the analysis of situated examples, the location and use for designed frailties as social individuation and care opportunities are identified. Finally, the controversy about the authorship status in tradition is scrutinised with an epistemological interest, and conclusions are drawn regarding the contribution of the design terroir notion to advancing an alternative diffusion strategy for mycobosci from the perspective of relational epistemology.

[51] Aesthetics is the philosophical study of the nature of beauty and taste, and is understood as the science of perception here.

4.3.2 Terroir

As a geographically situated cosmology articulated around a territory, terroir is one of the European heritages that could be built upon in an effort to develop sustainable systems of design production. This ecological and social quality, profoundly based on craftspersonship, ecological situation, and geographically situated psycho-social individuation (Simondon 1958), actually scopes beyond food and beverages. In 2005, INRA, INAO and UNESCO[52] defined it as the following:

> A terroir is a delimited geographic space defined by a human community that builds throughout its history an ensemble of distinctive cultural traits, of knowledge and practices, based on an interaction system between the natural milieu and human factors. The know-how at play reveals an originality, confers a typicity and allows a recognition for products and services that originate from this space and for humans that inhabit it. Terroirs are living and innovative spaces that cannot only be assimilated to tradition.
> (author's translation; Prévost *et al.* 2014)

In edibles, a terroir quality encompasses a complexity in consumer experience reflecting the peculiar and specific territory revealed through advanced craft. Fragrance, fleshiness, aroma, colour (*robe*), these qualifications can contribute to describe the pallet of experiences composing the sensory blend in wine tasting beyond taste. The essence of the wine, such as the history of the variety, the soil it is cultivated from, the age of the vines, or otherwise indirect organisational conditions of cultivation, play in the consumption experience and form a part of a cosmology of the

[52] INRA: Institut national de la recherche agronomique (national institute for agronomic research); INAO: Institut national de l'origine et de la qualité (national institute for origin and quality); UNESCO: United nations educational, scientific, and cultural organisation.

territory, of uncommons (Paxson 2010). And the uncommons meet the uncommons; the essence of the craft that composes the practice confers this aura or identity to the object (Benjamin 1935), while the rich experience of the consumer subsumes the bare functionality of the drinking of wine into an artful and *interesting* event (Evans 2022). This is generative of uncommons based on a different sensory experience between that of the consumer and the craftsperson's. It ties, in its co-becoming through tasting, the consumer to the artisan producer and the identity of the geography itself. Terroir is a resistive way of knowing, in contrast to an objectivist scientific explicitation (Teil 2012).
It is remarkable that the previous definition opens the scope of the qualification beyond edibles. In terroir, the environment is so associated and intertwined to the mode of existence of the object that the methods and materials are protected from modern normative food safety demands—with conditions of control of the finished product. The fauna, flora and microbial fauna are a core part of the design with instances such as usage of wood in ageing, or recourse to naturally occurring ferments. While the dependent existence of the products and services to geographic conditions is mostly inherited out of pre-modern logistical necessity, the fragility is nonetheless conveyed in the bills of specification for the products as a way to guarantee its persistence.

Reducing technical integration and entanglement by embedding technological dependencies to societies and ecologies—frailties—is equivalent to setting intrinsic environmental and social exploitation limits. In the French law that regulates the protected salers cheese for instance, it is specified that it should be produced on the milking site, from a list of villages, between April 15th and November 15th, and when cows are eating field grass (*Décret du 14 mars 2000 relatif à l'appellation d'origine contrôlée « Salers »* 2000). This is a regulation that makes it easy to identify the degree of locality of the salers design: the materials are highly constrained, both in time and space; the method and tools are open, detailed and regulated by law; the fabrication process must be manual. The mode of existence of the salers cheese therefore protects its environment while protecting locality by ensuring that the design is bound to, for instance, not integrating an unsustainable level of automation that would contribute to adding a distance between the craftsperson and the biome. Under the constraints of climate shifts and other environmental externalities, the fragile design may as well serve as a precursor sensor of these, like a canary in a coal mine.

A terroir quality refers to the co-existence of various cultural expressions and cannot be reduced to an isolated design. A design belonging to a terroir cosmology therefore best expresses this quality in its relations to other situated designs. In the case of the salers cheese for example, it situates along other protected cheeses: the *cantal*, *laguiole*, *saint-nectaire*, *bleu d'Auvergne*, the *fourme d'Ambert* and other, less considered ones but integral to the cultural landscape. This production relates different actors about the cheese production organisation: the producers and distributors, the political representations from municipalities up to the national level, and unions and associations (Brosse 2009). The Cantal area of salers production is also populated by vernacular constructions made of rubble stone and slated roofs, generally called *burons* (shed, in ancient French) borrowing from vernacular language. These small sheds serve to host farmers aestivating[53] in the heights of Cantal, and to host the making of the cheeses[54]. Furthermore, the going into aestivation, called transhumance, is the opportunity of a festive event for the local inhabitants to get together and walk along the herds to accompany them into their temporary graz-

[53] Aestivating means to accompany the herds to rich pasture for the summer period.

[54] Burons are not used to produce cheese anymore on a regular basis, and aestivating has fallen into desuetude too with the advent of new forms of food for animals and modern logistics (Brosse 2009). Transhumance is still celebrated nonetheless among other traditions by advocates of pastoral, non-industrial agriculture.

ing locations. Transhumance is a practice estimated to be 4 000 years old and is common to the mountainous areas of France (Biber 2010), Switzerland, Spain, Italy, Austria, Morocco (Auclair *et al.* 2013), and Eastern Himalaya (Barsila *et al.* 2015), to name a few. As the principles of cheese making are fairly simple and widespread, the specific protocol and tools influence their situated characteristics. Each terroir cheese has its specific tools, for instance to cut the curdled milk into blocks to release the whey and remove humidity to favour safe preservation. The tool design influences the size of the curds and the subsequent remaining humidity after pressing. As introduced at the very beginning of this thesis, the specific design of this humidity and potential creaminess, in combination with brining for instance, often relates to the geography, ease of distribution of the product, and preservation needs. Natural ferments are often used too to preserve and add somatic quality to the artefact. Their activity and presence may be be incentivised by specific cleaning and fabrication techniques (for salers, see: Bérard and Montel 2012; Callon *et al.* 2004; Duthoit *et al.* 2003). And the salers cheese can be even further characterised by the origin of its milk. When the milk only originates from Salers cows, the cheese is called *tradition salers*. The presence of this cow race in Cantal, along with the prototypes for salers and cantal cheeses, is estimated to be 2 000 years old (Kindstedt 2012).

This non-exhaustive review of the mode of existence of the salers cheese illustrates uncommons in excess, in the eco-social relationality that the design lives by. The excess can be valorised in contrast to a rationalised conceptualisation of cheese. If cheese making is to be rationalised as a method to store milk proteins during the colder months, simpler forms of preservation techniques, such as brining, may be more economically efficient than inoculating natural ferments such as *Penicillium camemberti* or *Penicillium roqueforti*. This would result in *mozzarella*-type cheeses, for example. The rationalisation and cancelling of the excess is also on dis-

play in the de-seasoning of industrial milks production with artificial insemination practices (Clarke and Knights 2022), thus questioning even the relevance of the rational need for industrial cheese if proteins can be available all year round. But cheese has a longer shelf life than milks and facilitates logistics, again.
There is no rational need, in modernity, for terroir-anchored cheeses and their fanciful excess of qualities. But their socio-geographically local specification reflects aspects of the communities' cosmologies, impacts individuals' social individuation about specific somatic experience in herding, crafting and experiencing, and influences a social conception of biomes and ecology (Brosse 2009; Paxson 2010). This starts to describe the cosmology of the terroir that salers refers to.
Terroir presents a millennia-experienced context of cultural geography, and is a quality that a design system may gain in the scaling of its bioregional relations. Although traditionally defined from a French context, it has gained a practical and academic interest for it "*offers a theory of how people and place, cultural tradition and landscape ecology, are mutually constituted over time*" (Paxson 2010). Cultural anthropologists Heather Paxson and Geneviève Teil warn in their studies of terroir about the potential for the commodification of the term nonetheless, much like organic labels, which may deplete the understanding and usefulness of the term (Paxson 2010; Teil 2012). "*Taste of place is not the same as taste of proximity*" (Paxson 2010).

Cosmology is an important idea in this context, because it situates the stakes in both the ontological *coherence* of the industrial entanglement (E. H. Giraud 2019), and the nature and amount of work ahead for the development of *coherent* alternative ontologies. Cosmology presents an intrinsic resistance to being studied too, as it can never be described as a whole given that this presents a risk of essentialism of the studied communities, and needs to be considered alongside other fields of study (Abramson and Holbraad 2014). The identification of the dynamics that influence it, and the description of technical landmarks that characterise situated case studies, can help to devise an alternative strategy for the democratisation of mycobosci. Before continuing the examination for the characteristics that may be identified in vernacular designs, a perspective is offered over the way terroir has been used as a regenerative method for international development by the UN.

4.3.3 Terroir in international development

Far beyond a romantic pastoral notion, terroir has been a key axis of economic development for the UN from the mid-nineteen-eighties onward, after a general movement away from projects emphasising technical aspects of rural development in Sahelian West Afrika. At a time when there was a rising recognition that the impact of agricultural intensification over natural resources and the environment had failed to be considered, *Gestion de Terroirs* (terroirs management) projects started to be implemented with three main approaches (Toulmin 1994; Dalal-Clayton *et al.* 2000):

- Natural Resources Management: focusing on the physical improvement of the natural resource base through existing institutions,
- Institution building: creating and supporting community-based institutions by thinking the organisational structures, economic incentives to manage, and returns gained from it,
- Local development: organising communities and the transfer of financial and legal responsibility for rural land use access and planning to the committees.

The method means to transfer the management of natural resources from governments to local inhabitants. Its implementation in Burkina-Faso as a Community-Based Natural Resource Management (CBNRM) has been successful enough to be adopted by the Ministry of Environment and Water for the management of the 10 most important forests and wildlife reserves in the country, and the approach was used to develop a national forestry policy in Mali. Although considered initially slow, complex and representing higher costs to other development strategies, the approach was qualified as the best available for addressing the socio-economic needs of the population and long-term land sustainability, by using local empowerment and capacity building (World Bank 1998). The inertia of governments in passing power to the communities, as they held onto the control of access to lands, confounded the responsibility of these programs and increased the inertia of pilot projects (Toulmin 1994). The Gestion de Terroirs approach has a strong focus on community-led local institutions and know-how for fostering both the autonomy regarding natural resources management while promoting resulting craft and cultural identity. The success of the approach may be related to the agency that was returned to the villages, and the main limit to the inertia in returning control from the states. Although the prime goal of Gestion de Terroirs was to limit environmental depletion and foster a regenerative dynamic, its eco-social design, as also paired with the restoration of local institutions, seemed critical to success. From a philosophical perspective, the inhabitants were invited to co-become again with their environment and mend their related uncommons, redeveloping a way of knowing that is situated about ecology. This series of projects further contributes to bring forth design alternatives to englobing industrialism and demonstrates, in a development context, that ecological resilience resides in the valorisation of intersubjectivity, local agency and democratic local institutions.

4.3.4 Frailties

Craftspersonship is central not only to the terroir, but also to vernacular architecture. Vernacular architecture traditionally relies on a distribution of the craft knowledge among the community to improve its resilience. Shared constructive methodologies that have been designed and tested by the community serve as community pre-individual knowledge[55]. These allow individuals to be able to both design their works with confidence in their safety, as the knowledge is inherited from tested designs, and to perpetuate the reproduction of design characteristics indicative of a social belonging. The materials that are used in vernacular architecture are traditionally subject to rather rapid erosion depending on the location and traditions—burons rubble stone works would be sturdier, while the widespread quality of more fragile clay also makes it into a versatile vernacular opportunity. The shared craft know-how and historical experience in maintenance can form opportunities for ritualised caretaking of the works, bridging the gap of relative fragility with community perpetuation. Furthermore, the definition of terroir that was cited earlier does comprise such practices: the geographic space is defined by a community production based on an interaction system with the natural milieu and human factors. Terroir is the quality of a design that depends on both these factors: it functions if the ecology and the human organisation around permit it. In this context, the resilience of the community is as great as its technological fragility[56].

As an example drawn from vernacular architecture, the Great Mosque of Djenné, initially constructed before 1330 out of rammed earth, illustrates what an architectural terroir may look like. Found abandoned in

[55] Pre-individual knowledge is that which pre-exist from the individual experience and has yet to be learnt from and adopted comprehensively by one (Stiegler 1998).

[56] This should not be mistaken for precarity, for it is evident that minimal knowledge access, know-how valuing and material accessibility are a requirement for fair and just living conditions.

the nineteenth century, it is after its reconstruction in 1907 by French colonialists—supposedly faithfully to the original design (Leiris 2020; Bourgeois 1987)—that the mosque has gained a great level of interest for its population. It is unique for its infamous yearly participatory replastering that mobilises most citizens. Because sun-dried earthen plaster wears off rapidly given the environmental conditions of Djenné, it offers a frequent opportunity for care enactment with local resources (Brunet-Jailly and Scherrer 2018). Virtually any clay could be used as a plaster; the local population sources its raw material from the floodplains of the river Bani, just outside Djenné. The design of the mosque makes it dependent, not only of its geography and climate, but also of the social structure around. This organisation, reflecting in efforts to establish local and autonomous institutions in Gestion de Terroirs, sheds light again on the reason for demonstrated resilience of terroirs: its practical and ontological entangling in people and environmental non-humans rather than seeing a purely rationalist delegation of management power.

Designed impermanence can also be based on other methods than wearing. The Shintoist tradition of Shikinen Sengū-sai (式年遷宮祭) consists in rebuilding temples every twenty years according to original construction methods and materials. The temples are known to be rather ornamental and sturdy; the process of reconstruction is a celebration of continuity through reproduction rather than conservation (Coulmas 1994). Here, the introduction of an impermanence via the ritual both supports a specific religious ontology and, as for what concerns the present study, community fostering by performative social individuation.
Fragility can be a parameter to balance the design of a technology (or of an architecture) and its dependency to humans and ecologies. Introducing fragility into a technology is a way to shift the power, from the author of the black-boxed knowledge that is embedded into an integrated design, towards collective intelligence. This intelligence reflects in the develop-

ment of crafts and somatic tacit knowledge (Ingold 2001; Collins 2010; Polanyi 1958), supported by explicit knowledge as semiotic conveyors. This is illustrated in traditional vernacular architecture with oral and written traditions, but also in agricultural terroir as discussed earlier. The building with rammed-earth or the development of a terroir, although it might result in impermanent designs, confers an agency to the community in managing the natural resources and incentivises a frequent care or *return to the object*. This invites for the increase of a technological autonomy from the need to acquire, develop and retain somatic tacit knowledge within the community. This opposes an expert delegation via the multiplication of interfacing technologies and institutions (Gorz 1992; Illitch 1981). The development of this local technological capacity contributes to form a terroir by reducing the distance from the user to its environment, and by making this sensible relationship a non-negotiable trait of the mode of existence of the technology. Paradoxically, the autonomy of the community is acquired through strategically placing its actors into a deeper dependence to each other from a relational perspective. If the work is not able to stand ten years worth of environmental wearing, then the number of educated people on maintenance that are available should make up for it, or the social organisation reflect it more generally. This suggests that these frailties, those weak points, are and can be implemented strategically to stir the structuring of a community constitutive of the design such that they co-become.

4.3.5 Tradition and authorship

A turn to locality and relational design, as learning from terroir and vernacular architecture, may invite for a critique of the supposedly conservative nature of tradition, and the potential lack of self authorship.

Tradition's detractors associate it with stasis and contrast

> it with change, but it is rooted in volition and it flowers in variation and innovation. (…) [It] opposes the alien and imposed.
> (Glassie 2000)

This mention of "*alien and imposed*" by Glassie can be set in perspective with Francia Márquez' letter excerpt presented in section 4.2.6, and relate it to a desire of epistemological autonomy. While modernity seems to ideologically oppose the modes of existence of traditions, the two notions can co-exist in one and communities to various extents (Galland and Lemel 2008). In the context of cosmological influence or colonisation, Abramson and Holbraad note that:

> while for many anthropologists the indigenous cosmos will forever be a largely powerless fictional representation, [the new anthropological body of work interested in cosmology] connects with cosmological openness by to illuminate the plethora of actual ways in which foreign forms (e.g. mines, money, medicines, white persons, white gods) are not only incorporated into customary 'ways of life' but are radically re-signified within and through existing cosmological frames (…).
> (Abramson and Holbraad 2014)

The authors also challenge here the fixed and closed idea of cosmological description, and identify the risk of exotising different cosmologies than that of the observer's.

Vernacular, traditional or folk designs all have a strong common trait in their mode of production: authoring is humbling as inevitably diffuse because of their shared nature (Bronner 2005). The self-construction and vernacular in the sense of situated and pre-modern, or non-industrial, challenges the authority and exertion of power from a single individual or community. In tradition, the creative act is collective. The individual-originated token contribution to the cultural ensemble makes use of pre-individual knowledge for multiple reasons. The first, and maybe most important, relates to learning in safety; to obtain a dwelling which stands and is reasonably durable, the institution provides axioms of sound constructive technique, much like an encyclopedia of edibles and ways to process them for a sustainable diet. When cooking a learned recipe, people can amend the design on-the-spot given the extent of an understanding of situated and sound relevant possible additions in time and space (to topography, resources at hand, climate or seasonality, for instance). This is more than functional prosthetics: the pride and humility that emerges out of the repeated enactment of this object reanimates *uncommonable* souvenirs of either or both elements of a natural milieu or the spirit of a relative (Pitrou 2019). This is a second interest for pre-individual knowledge in tradition. The pride is in inexact similarity, inexact replication, as they symbolise a belonging and intimate valorising of milieu while the local adaptation of the design also signifies people involvement in adding a stone to the heritage in tradition. Lastly, this contributes to situate oneself in a context of shared power, both politically within the community, and ecologically with regards to non-human beings. Situated designs act as proxy to a shared memory, and are thus fundamental in a renewed social individuation (Stiegler 1998).

Simon J. Bronner exemplifies the relationality in tradition with the Amish barn raising. A Herculean task for a single person, the tradition of community building of a barn in this culture involves negotiations and the repetition of reciprocal help. With all the tacit expertise of the peers, the family raises not only a building for themselves but a host to future social gatherings such as religious services (Bronner 2005). While a more rationally technicised and market-supplied solution, such as delegated to a construction company, would provide the family with more available time, this tradition ensures a durable resilience beyond the project. Non-industrialised architecture in this sense is intelligent about the retaining of tacit knowledge within its corps, and ultimately fair about its material and human mobilisation. Fair, because in contrast, the technical delegation through functional entanglement distances the intimate and collective sensibility from local biomes, and naturalises a positivist epistemology of nature.

4.3.6 Relational epistemology

The three aspects of safety, social belonging and ontological situating are based on pre-individual knowledge and their affordances. This knowledge, present before the individual comes to the society, is a continuation of the collective memory awaiting acquisition by one. This *non-lived already there* carries a lot of influence: it is at the same time the receptacle of the memory of its production methods, reflecting human learning from experience, and what results from the production methods with the archetypal existence of the knowledge (Stiegler 1998). The knowledge is both the result and the condition of its production. As a pre-individual knowledge in potential, the conceptualisation of a design influences the possibility of a world. And all the more when it bears a lasting material form. The design as a knowledge carrier interfaces between the individual, a person, and a community such that the person, community and the technique are defined in intersubjectivity. This co-defining is essential to the ontological coherence of a situated society, and is greatly influenced by geographical conditions in pre-modern contexts. Nurit Bird-David sums it up: "*Against* 'I think, therefore I am' *stand* 'I relate, therefore I am' *and* 'I know as I relate'" in a relational epistemology (Bird-David 1999). This inseparable and relational understanding is elaborated upon by Stiegler with regards to knowledge naturalisation:

> The *Dasein*[57] inherits of the already there as a past that always preceded it and from which it is *that* individual, son and grand-son from such-and-such, etc.—*their* past which is nonetheless not properly *their* past, as they have not *been lived*: the temporal mode of being of the *Dasein* is the historicality that is always already affected by the facticity of this *non-lived* past where grounds the banal understanding of self.
> (emphasis in the original text; author's translation; Stiegler 1998)

Pre-individual knowledge is further characterised as a *positive facticity* by the author, because it allows for one to have an access to their historicality and situate ontologically. The implications of this facticity in knowledge transmission range widely on the philosophical domain, and will not be studied presently to the benefit of cohesion and clarity of the study. The qualification of facticity suggests the possibility of a naturalisation, particularly in the context of commodification.

From the review of relational individuation elaborated by Stiegler, it appears that a Western epistemological tradition that valorises pure cognitive productions in isolated individuals[58] (Thayer-Bacon 1997) is reductionist at best, appropriative at least, and probably both as per uncommons. The relational view bears significance for the relationship to biomes: how can a community decide on the democratic closure of a negative common, while the knowledge about its technicity is shielded, unverifiable, the service it provides commodified, and one cosmologically conditioned not to have the ability to identify the affordances of the commons even? The retention of knowledge in centralised institutions or companies hinders the possibility of a democratic intervention, of a technical autonomy, but

[57] Stiegler qualifies Heidegger's *Dasein* of an individuation (Stiegler 1998).

[58] See also Alfonsi 2019 about the persistent masculinist conceptualisation of the artist as an isolated genius, with regards to authorship.

also of the possibility for the ontological auto-determination—or individuation. Uncommons as qualified in section 4.2.6 are a product of relational individuation, and bear the ontological importance of historicality for one.

The production and use of objectivised knowledge surrounds societies in various contexts, from linguistics to mechanical standards, through evidence-based medicine practices to economic transactions and legislation. Their character of "*general anonymity*" leads to the durability of knowledge and overrules personal opinions (Thévenot 1984). The attaining of a scientific consensus may lead to a normalisation, or standardisation of new knowledge thanks to the use of scientific methods supposedly leading to objective information (Thévenot 2009). But objective knowledge without praxeological situation (Bourdieu 1973), i.e. too englobing a normalisation or the lack of affordable methodological demonstration, may lead to the challenging of the legitimacy of consensual objective knowledge (Beck 2011). After reviewing the interests and backdrops of standardisation practices which lead to naturalisation, sociologist Laurent Thévenot states:

> And yet the treatment of plurality is at the heart of politics. To celebrate time saving and alleged value free procedures through standards is to forget that explicit politics have been subsumed into the elaboration of the standard objective, often removing them from any opportunity for open political debate.
> (Thévenot 2009)

While the use of scientifically consensual forms of knowledge is inextricably important for Western sociability and politics, as much as for other forms of societies using normalised pre-individual knowledge, it also enacts power relations which may institute a lasting asymmetry between

stakeholders (Castel 2009). This asymmetry is an unequal exchange.
It can be illustrated by the case of mycoboscus design: if industrially produced, standardised mycoboscus compositions are commodified, they are normalised for economically rational production and distribution. It was demonstrated before that this model implies not addressing the ecological risks that the technique involves, making it not absolutely sustainable and risky. Moreover, by having recourse to commodified products, one naturalises the power that is commercially imposed upon them, and naturalises a lasting inconvertibility of the relation. In the present case, standardisation and its interest is distinguished between imposed and negotiated. In an economic context, for consumers to be able to "vote by buying"—if this concept has ever any political relevance—they need both access to competitive alternatives (in a red ocean) and knowledge about the product or service. These conditions are not granted in the present case context, for example. The consensual agreement conditions to reach a sound standard are violated, such that citizens are implicitly required to consent to using ecologically risky products, and to accepting not to know what the technical description of the products involves. Industrialisation and commodification, here, crystallise a power relationship in an unequal exchange of compromises, to the benefit of capital accumulation although the industrialist presents mycobosci as sustainable.
While the crystallisation of knowledge as scientifically rationalised and objectivised, meaning its naturalisation, can be effective for Western development and political life, the possibility of its scrutiny and democratic acting upon is impacted by the conditions for the democratic access and maintenance of knowledge about in societies (Thévenot 2009; Thévenot 1984). The possibility of a democratic maintenance of knowledge is then a possibility for the democratic maintenance and adaptation of cosmology. The objectivisation of unequal power relations should be the best advocate for supporting the possibility of a democratic review and amending,

in the context of sustainability and social justice (Dorninger *et al.* 2021; Hickel *et al.* 2022).

Safety, social belonging and ontological situating are some aspects that a situated community can contribute with, but it hardly distinguishes a Western objectivising society from an animist one. This is a great news, to avoid the caveat of dualism and for the prospect of the relational project in a highly expert-mediated Western society. Relational epistemology helps to identify the significant influence of technology on our ways of knowing and being; this suggests a heightened power and responsibility in designers as practitioners of *applied arts*—the synthesis of technique and arts. As such, designers need to be concerned with epistemology to devise their production and diffusion strategies: identifying and analysing (negative) commons from an apparent issue, proposing alternatives that are not technicist but leverage the geographic and social dependence, and anticipate negative effects by allowing for the impermanence of their designs and its amenability by the greater number.

4.3.7 Conclusion

The desirable local development of the mycoboscus technique as affordable and aware of the inherent ecological risks it presents also demonstrate social development opportunities. While a centralised industrial diffusion of mycobosci uses may be praised for its supposed efficiency and speed, it is worth noting that the volumes of the non-biodegradable and non-recyclable expanded and extruded polystyrene (EPS and XPS) materials production have been increasing since 2007 nonetheless. The global EPS market has seen a 1.5-fold growth in turnover between 2000 and 2010 (Future Market Insights 2022) and is expected to grow by 5.68 % over the 2019 – 2028 period (Triton Market Research 2018) or 4.7 % over the 2021 – 2028 period (Grand View Research 2020). The global XPS production is expected to grow by 4.0 % over the 2019 – 2024 period (MarketsandMarkets 2019). Fifteen years after their industrial diffusion began, mycobosci have still not significantly contributed to reduce plastic use, although the principal actor of this dynamic is fiercely trying to defend a market and knowledge monopoly.

Should it be desired to develop and implement mycobosci as sustainable towards market-readiness, locality will be critical here again. To constrain a design to a set local context, the technology that supports a design can embed frailties related to its local bioregional environment. These are important in that they often depend consequently on the social organisation about. Or it can even be observed that the social organisation may actually be a prime object of interest for designers as the case of the Shikinen Sengū-sai illustrates. What appears clearly in the traditional mode of existence of the examples that were mobilised here, is that the social individuation revolves not particularly in the act of technical maintenance, or impermanent mending of the frailties. It rather anchors in the maintenance of the knowledge and its transmission. Both in Amish barn raising, Shikinen Sengū-sai or the re-plastering of the Great Mosque of Djenné, the retention, re-actualisation and introduction of a community tacit knowledge to newcomers is foundational to an ontological autonomy and belonging. This knowledge that is integral to the community cosmology situates in the development of a social and intimate uncommon. It can further be deducted that the epistemological colonisation from Western objectivism, accompanied with its naturalist ontology of nature (Descola 2014), has negated this critical aspect of epistemological justice, and in the same arc deprived most societies of their capacity for socio-environmental individuation and sensible environmental care. This is a very consequential effect of expert mediation, typical of industrialised societies, that further invites for a revision and extension of the studies about this mediation to the possibility of an environmental regeneration (F. Fischer 2000)[59].

In the terroir quality, the design is both geographically and socially dependent. As proposed before, it might even just be that the bioregional constraining actually resonnates for its social belonging, its contribution to self-situating. The notion of dependence for a technological specification is not to be confounded with geographical exploitation in a *resource*-relationship. Developing a terroir approach as bioregionalist and fragile is one of the relevant democratisation paths forward for the craft and use of mycobosci. The geographical distribution, diversity and versatility of the wood-rot fungi considered for use make it particularly affordable, and the base knowledge for developing minimal valuable design functionality presents a very low barrier to entrance. There is thus no particular interest but that of commodification to favour a monopolistic delegated industrial capacity, in the context of mycobosci. As commodification results in what could be qualified as negative uncommons—the naturalisation of a distance of intimate sensibility to biomes—it is recommendable that the industrialisation process is challenged.

The need for reconsidering alternative epistemologies favouring subjectivisation also points to the development of new aesthetics in potential. Terroir relies upon an intersubjective co-becoming based on the excess sensible experience subsuming bare functionality. This experience is in itself a somatic tacit knowledge transmission method, reflecting an excess design specification—from the base logistical need to store milk proteins—that contributes to give a fragrance, fleshiness, aroma and *robe* to the territory. Terroir has a qualitative interest rather than a quantit-

[59] Many design practitioners and theorists identify social individuation as critical in their eco-social practices. The development of eco-social design (Elzenbaumer *et al.* 2017), bioregionalist design (Rollot and Constant 2018; Papanek and Lovejoy 2022), ontological design (Escobar 2018), relational design (Maier and Fadel 2009), designerly autonomous methods (Friedman 2003; Brand 1995), restorative design (Toews 2018) and *design for sustainability* (Spangenberg *et al.* 2010) exemplify this coherent recognition of situatedness as critical of expert mediation for environmental care and social equity.

ative, scalable one, and this was appreciated by the UN while developing the quality into a method for international development. The focus on local institutions building that they adopted reflects clearly on the main keystone: the social individuation of its inhabitants around a landscape. In scaling down the governance of the land towards specification and proximity, further developing the livelihood support capacity of the environment for villages, it allows for a fairer commoning. The asymmetries of power that may exist are acknowledged, between a village and a national government for example, and addressed to improve the maintenance quality of the environment. Doing so reduces the reductionism inherent to the globalisation of a managerial power that needs to consider multiple contexts systematically with a bird's eye view, and facilitates the negotiations in commoning as limited to the stakeholders directly dependent and sensible to the landscape. The process of commoning aware of scales and power relations may as well facilitate the development of a local uncommon, a local cosmology that is not imposed from a distance.

The inexact replication in craftspersonship education and practice further contributes to grant agency to situated communities. The affordability of a system allows for the collective, potentially democratic intervention on the design. Improving design technical affordability, by opening the blueprints and limiting technical integration, and supporting their adoption by a form of craft transmission is critical in a world of rapidly evolving biological states. Any developing institution or infrastructure should integrate a backdoor to its design allowing a public and scientific evaluation of its sustainability and social effects, and be re-engineered or dismantled should they be judged to have become a negative common. Introducing frailties can represent this backdoor, and depending on the nature of the design at hand, this needs to be designed according to the specific technology or system. This, will allow for the possibility of a democratic commoning. The ritualisation of a maintenance, or dismantling and rebuilding even if the design is sturdy, can be a way. This can contribute to de-naturalise the previously commodified and increase the ontological coherence of a community in subjectivisation. The granting of epistemological affordances to a general audience, or citizens, conditions the possibility of a democratic maintenance and change of people's cosmologies.

As knowledge is identified to be the critical unit here for eco-social design development, this presents an opportunity to study further the effect of knowledge carriers on the adoption and development of tacit knowledge with a view to facilitating citizen appropriation and ownership. The notion of controlled equivocation proposed by Viveiros de Castro is of a particular significance here. The somatic tacit knowledge is typically acquired by apprenticeship and replication, until movements flow in and out (Ingold 2001). No gesture of the craftsperson will and can be the same, both in persons and time. Body differences, one's cognitive and sensible history or heterogeneity in the material supply such as in woods, and many other specifics help perceive the craft activity as based on controlled equivocations. From sensible principles formulated by an experienced teacher, the apprentice typically means to develop their own sensibility, gestures, and their artistic expression in necessary corporeal equivocation. Furthermore, this is based on a rich sensory blend involving all senses—caressing the wood grain, appreciating the warm smell of sanded wood, the sharp abrasive sound of the blade, the rewarding curvatures of the long worked design. The case of know-how illustrates well the intent of Bourdieu with the naming of praxeological knowledge as situated objectivised knowledge (Bourdieu 1973). Anchored in praxis, it is meant to critically make sense for one of pre-individual knowledge in the process of sensible embodiment and individuation.
Architectural education inheriting from a fine arts tradition valorises the synthesis skills of the designer as a tacit or praxeological knowledge,

for example by granting master programs a considerable time for studio and project works in an apprenticeship model (Gaudillière-Jami 2022). Explicit knowledge carriers are ubiquitous to pedagogical activities and architectural technologies training. Their pedagogical efficacy was evaluated in comparison to somatic tacit knowledge activities in the context of mycoboscus craft teaching activities, with a view on its interest for general audience dissemination.

4.4 Epistemological experiment

4.4.1 Introduction

The development of novel material systems with a higher vernacular potential, like mycobosci, can help to practically and cosmologically address negative uncommons that persist in innovations deemed sustainable. The establishment of a mycoboscus scientific description and recommendations in section 2.12, and the identification of the central role of epistemology on social individuation and environmental care in section 4.3.7, indicate a need for the evaluation of the effectiveness of different knowledge forms for acquisition and development of tacit knowledge.

The diffusion and teaching of mycoboscus knowledge to applied arts students offers a unique opportunity to study a pedagogy that focuses on craft and tacit knowledge for its relational capacity. This study finds a more general interest in exploring the conditions of knowledge dissemination to an audience of amateur practitioners. As discussed previously, in this opportunity lies the potential to support an alternative ontological intersubjectivity within both the environment and society. Incidentally, the strong aesthetic presence of the practice—by the effect of submitting the senses to wood fermentation aroma (resembling that of forest humus), dense mycelia of a slimy and elastic haptic and its visual expression as *gross* (Fig.4.59, Fig.4.72 and Fig.4.61; Rigobello and Gaudillière-Jami 2021) or soft, textured, darkened and velvet-like (Fig.4.63 and Fig.4.62)—invites for a reconsideration of the role that somatic tacit knowledge play in architecture and design activities. In her analysis of the computational design field, Nadja Gaudillière-Jami argues that: *"The role of digital tools in the pedagogy is a means to interrogate what students learn about architecture itself, rather than a technical stake."* (author's translation; Gaudillière-Jami 2022 p.273). Hardly reducible to sole mechanical properties or abstracted analysis, in fact both the mycoboscus craft and the experience of a finished mycelium material demand a bodily presence to be fully appreciated. In activities tending to favour computation-based works and thinking, explicit knowledge has progressively been made central to the pedagogy, along with expliciting activities, so as to render a design brief computable. This reflects a progressive turn to the use of commodified objectivised data in this practice, an epistemology that favours phenotypical expression in its positivist heritage, but discards the excess possibility of life. In a study of the contemporary history of the computational field in architecture, Gaudillière-Jami has furthermore reported that with the succession of generations of practices and a democratisation of computational design, the acquisition of coding tacit knowledge has generally become impoverished because of the structuring of commercial software solutions as centralised tools rather than sustainable open source practices (Gaudillière-Jami 2022 p.410). Gaudillière-Jami elaborates on the impact on practices, where newly trained architects would tend to be subjected to centralised software biases and reductionism, and naturalise these as the detailed operations of the digital tools are obscured for them. It is therefore important that the pedagogy supporting mycobosci knowledge transmission presents the opportunity of working with a living fungus not as an objectivised technology, but as a praxis aiming at being generative of a tacit ecological sense.

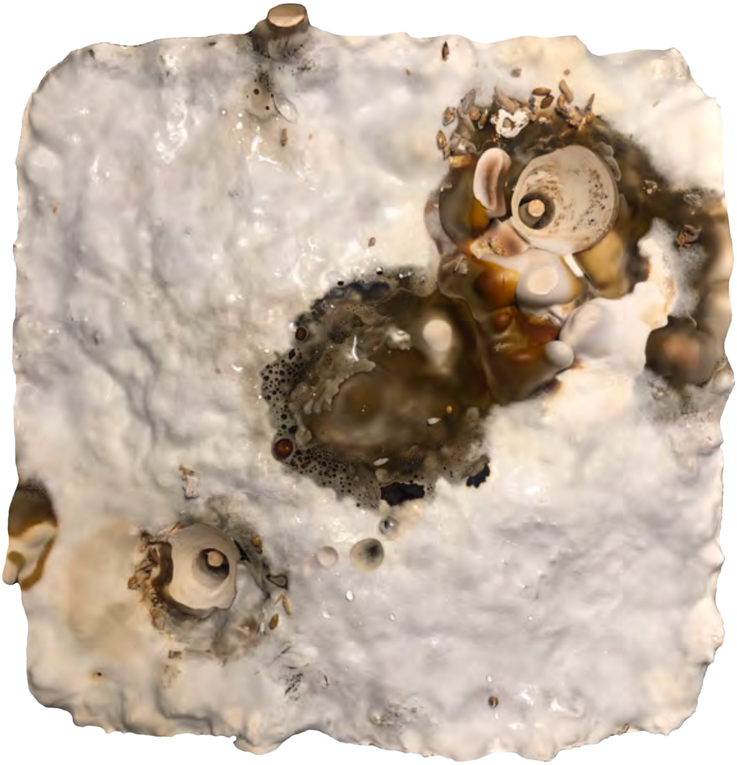

Figure 4.59: Top view of an experimental sample (2020).

Figure 4.60: Healthy *G. lucidum* mycelium cultivated on beech wood (2022).

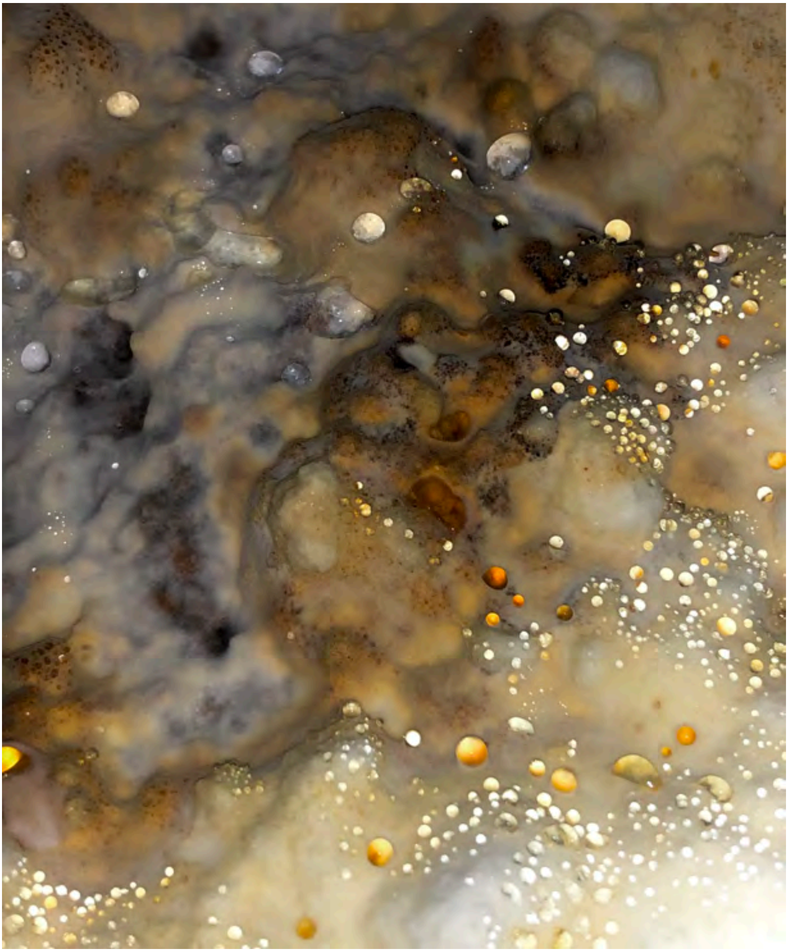

Figure 4.61: Detail of a healthy *G. lucidum* mycelium cultivated on beech wood (2022).

Figure 4.62: Ripened *G. lucidum* mycelium soft texture. Own work in collaboration with Joanne Jones (2022). Picture: Joanne Jones (2022).

Figure 4.63: Top view of ripened mycobosci (2021).

4.4.2 Liquid Modelling workshop

In September 2020, a team from the Fungal Architectures working group in CITA conducted a two part workshop as an introduction to the craft of mycoboscus to first and second year master students of the MA Computation in Architecture program, Royal Danish Academy[60]. Over a four week period in total, a first two week workshop called *Shaping Growth: composing material heterogeneity* was led by Phil Ayres and consisted in an introduction to the cultivation principles of mycobosci, cast production, composition of substrates with macro structures such as straw fibres and implementation of environmental sensing to map onto the digital modelling of composites. I led the second workshop called *Liquid Modelling: navigation beyond vision* that leveraged these previous works with the integration of a simulation model based on moisture dynamics and its influence on mycoboscus production. Gaudillière-Jami and I used this context to study the acquisition of tacit knowledge relative to mycoboscus craft and ontology by the students. For this, we conducted interviews with all the students at the end of the workshop. The simulation model, called *Prototaxites stellaviatori*, was designed specifically for this teaching activity and made to echo theoretical matters in mycobosci. Because of the centrality of the Rhinoceros 3D – Grasshopper modelling environment to the master program, the simulation model was designed as a plugin for this.

4.4.3 *Prototaxites stellaviatori*

Fungal colonisation is driven by a wealth of factors including moisture qualities and dynamics in the substrate, the fitness of the substrate chemical profile to fungal species enzyme array, the aeration of the substrate during fermentation, the UV stress, the presence of other microorgan-

[60] Part of the following of this text until section 4.4.5 was the topic of a research disseminated in Rigobello, Gaudillière-Jami and Ayres 2022; appendix F.2.

isms and the protocol care by the producing person. The simulation model focuses on integrating evaporation and capillary action as prime vectors for predicting fungal colonisation, because the presence of capillary water in solid-state fermentation media is a prerequisite to allow extracellular transport of metabolites in fungi and to establish osmotic potential for allowing turgor pressure and subsequent hyphal growth (Money 1995). It can also be noted that fungal decay by oxidation frees water along with CO_2 (section 2.6); this is not a phenomenon that was considered for modelling in this context. Lastly, the effect of water activity upon turgidity and enzymatic activity was not modelled. The simulation model can be used with hypothesising these non-modelled experimental factors as fixed, and can be used to explore the remaining of the design space allowed by the parameters.

Based on a discretised volume represented by a point cloud, the simulation is made of two parts: firstly the solving of moisture dynamics (evaporation and capillary action) in a substrate medium including structuring fibres, and secondly the solving of a mycelium development based on the pre-computed moisture dynamics. Because many factors other than moisture presence can influence mycelial colonisation, and because the physical model rendering may not be absolutely accurate, the simulation was made stochastic. Each data point has a confidence level affected for the three input datasets: moisture distribution map, warmth map, fibres location.

To illustrate, a user can set the warmth that they observe on the external boundary of an experiment with a maximum confidence level (1.0) as they can measure the temperature accurately; if they cannot directly measure the temperature at the centre of the volume they can set a lower confidence level. The probabilities are then being solved as the interaction of the events at each point of the model during the solving. This simulation model thus focuses on moisture dynamics, macro composition of fibres

and unpredictability.
The aim for this as a pedagogical instrument was to make it so it promotes a reflexive craft-oriented practice in students, an active dialogue between simulation as explicit knowledge-based activities, and a somatic tacit knowledge sense-building of working with fermentation practices. Beyond favouring a creative use of fibre placement such that it leads to complex spatial mycelium configurations, the model aims primarily at questioning the control that the designer exerts on a living microorganism towards mitigating the controlling of their expectations in formal negotiation. Illustrations of simulations results are presented in Fig.4.71 to Fig.4.74.

4.4.4 Evaluation

Using the SECI model[61] (Nonaka *et al.* 2000), Gaudillière-Jami and I evaluated the interview results with regards to the student productions (reported in Rigobello *et al.* 2020; appendix C.1). The conceptual density of the workshops for new comers to mycoboscus craft and computation methods was acknowledged, and the notion of stochastic modelling was less appropriated, in the reports from student, although the multiple craft iterations that working groups could perform contributed to the appropriation of the notion of uncertainty. As the four SECI conversion modes were identified, both in the crafted objects, in the reports and in the interviews discourse analysis, the learning hypotheses for the workshops were

[61] This model allows for the identification of four modes of conversion: *socialisation*, enabling a tacit to tacit knowledge conversion, and referring to moments of observation and imitation; *externalisation*, enabling a tacit to explicit knowledge conversion, and referring to explicating knowledge in writing or drawing, for instance; *combination*, enabling an explicit to explicit knowledge conversion, and referring to the use of existing explicit formats in order to produce new explicit ones; and *internalisation*, enabling an explicit to tacit knowledge conversion, and referring to the acquisition of tacit knowledge through the manipulation of explicit formats (Rigobello, Gaudillière-Jami and Ayres 2022; appendix F.2).

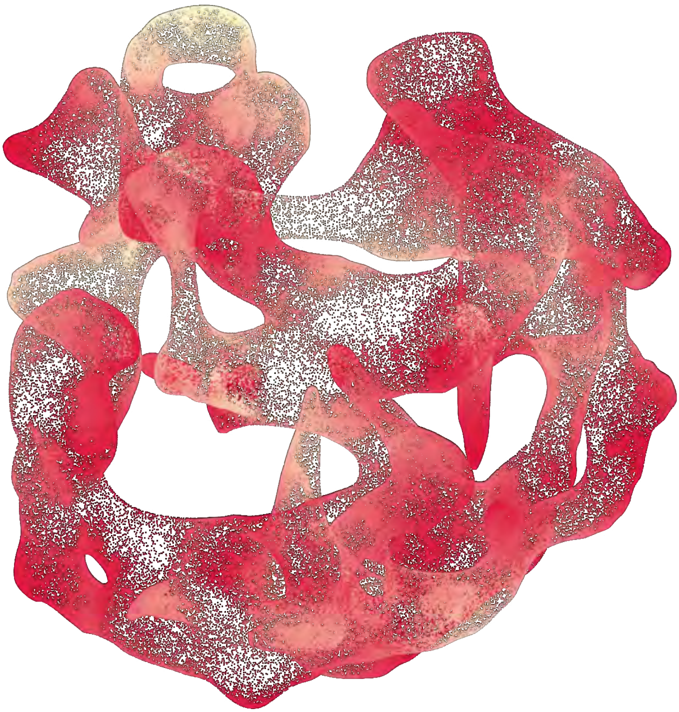

Figure 4.64: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

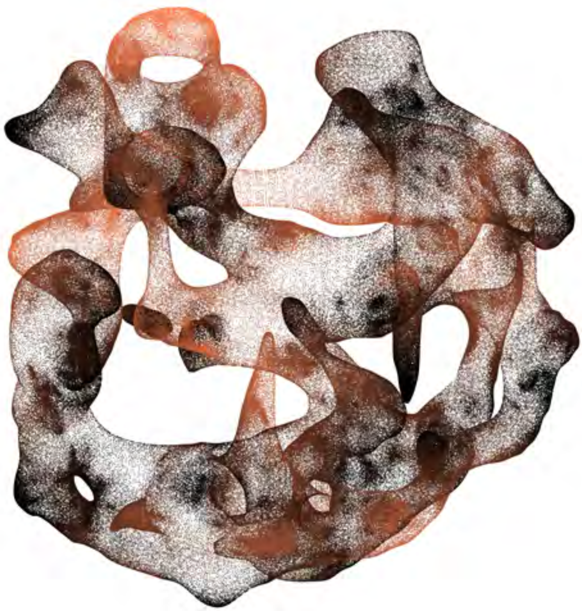

Figure 4.65: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

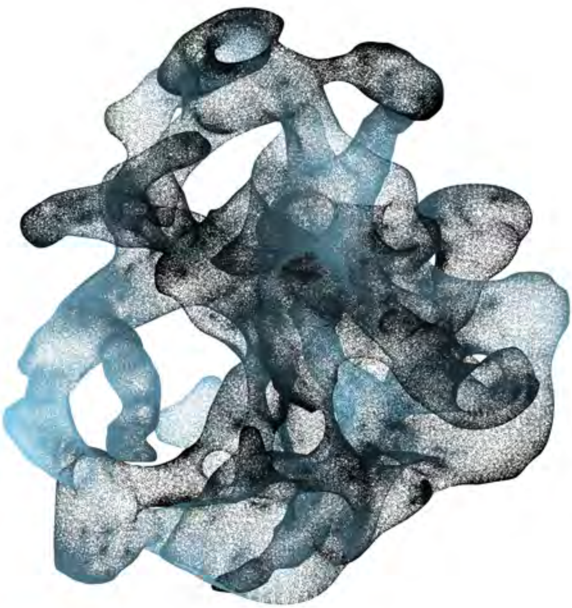

Figure 4.66: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

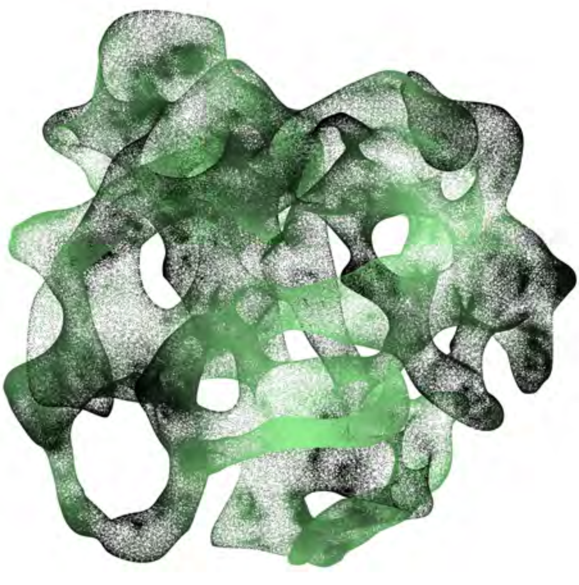

Figure 4.67: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

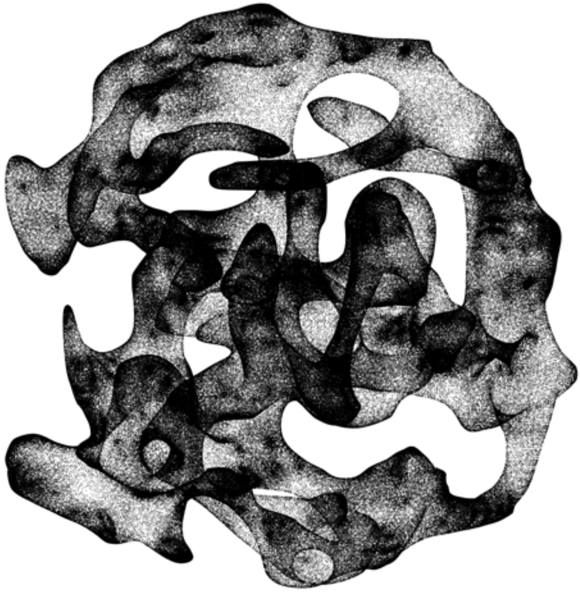

Figure 4.68: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

Figure 4.69: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

Figure 4.70: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

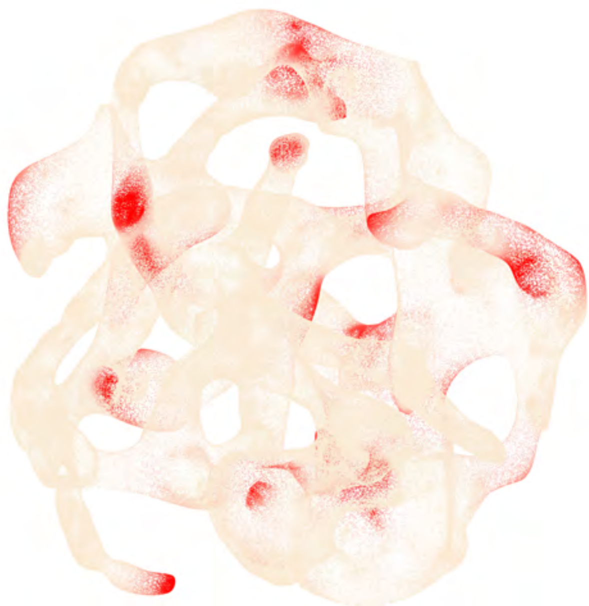

Figure 4.71: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

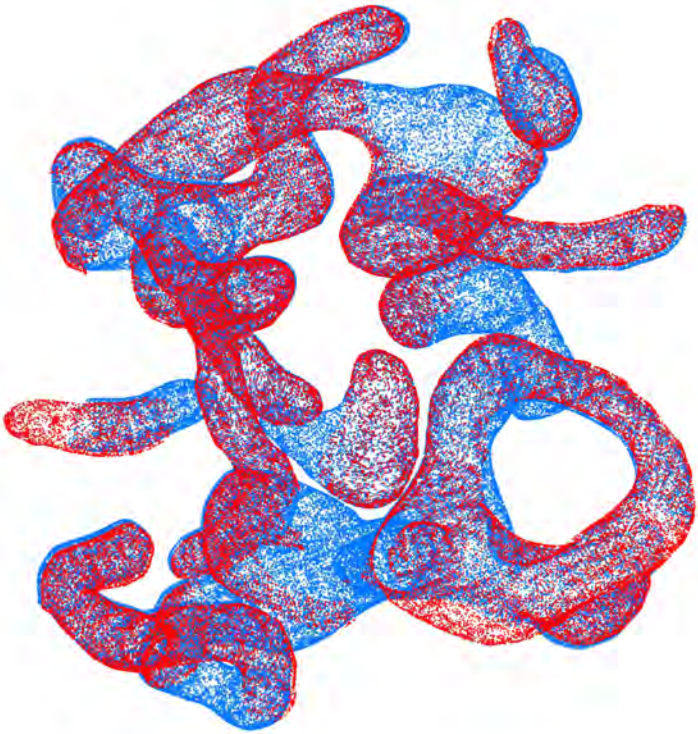

Figure 4.72: Fungus fuga. Edited simulated mycelium growths; colours are based on stochastic values (2020).

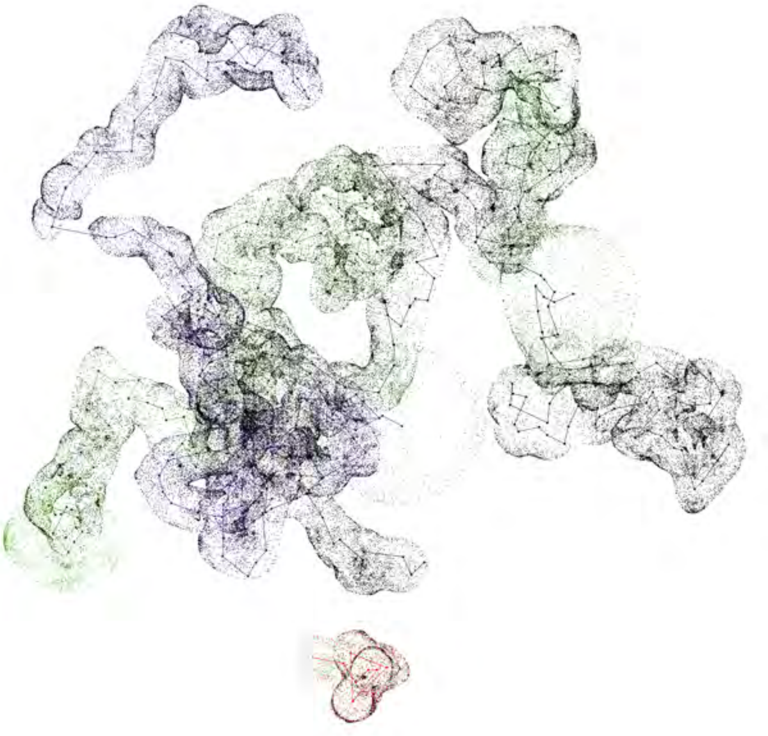

Figure 4.73: Edited simulated mycelium growth (2021).

Figure 4.74: Top view of valence two branching growth modelisation in Gaussian referential (2021).

corroborated as somatic tacit knowledge of mycoboscus craft was distinct and critical thinking about simulation model uses in this context was displayed (Rigobello, Gaudillière-Jami and Ayres 2022; appendix F.2).
The general lower appropriation of the stochastic capability of the simulation tool by students, suggests that the knowledge explicited and embedded into the model was not made affordable enough. Conversely, the somatic tacit appreciation by means of craft experimentation, led to students appreciating this uncertainty and humility in the manufacturing activities. The simulation model was presented as a digital tool packaged into a plugin with non-accessible code, and although it had been presented with tutorials and designed to be affordable, it could only hardly convey the notion of uncertainty as this required basic notions of statistics, computation logic and computational geometry along with a preliminary tacit understanding of mycoboscus craft to appreciate modelling decisions such as the moisture focus.

The craft activities were introduced as a soft apprenticeship environment with punctual help in the making. The latter allowed for a controlled equivocation favouring the appropriation for oneself by direct somatic involvement, while the use of a partially closed simulation model illustrated an inverse pedagogical method that was strongly guiding and necessitating preliminary knowledge that could not be assumed from a diverse group of first and second year architecture students. In this context which can be qualified as *expert mediated*, the students were implicitly required to perform a leap of faith as of the background knowledge which hindered their capacity for appropriation.

4.4.5 Conclusion

The mobilisation of biological methods tending to a scientific expertise in an educational context, where students are typically meant to be trained into a generalist design focus on synthesis and sense-building, provokes a productive reflection on the articulation of explicit and tacit knowledge carriers in dissemination exercises. Much like scientific knowledge of fermentation is not required to produce bread or pickle either as an amateur or a professional, the transmission of mycoboscus knowledge can be made very affordable to generalist profiles—and anyone—as of its minimal necessary knowledge for controlled equivocation. This content can be communicated with minimal explicit set points, for instance the initial moisture content of the substrate or the incubation temperature and duration, and leave room for both the somatic tacit experience and social individuation to unfold. This is especially relevant in the case of mycobosci, as the scoped fungal species allow for the use of a great variety of cellulose-based substrates. The usage of specific explicit knowledge beacons can help structure an equivocal process. The considered design of these nudges finds relevance again as the ontological situation needs geographical and community specification to local contexts to actualise its sustainability and social development potential. That is to say, the dissemination of completely explicit recipes may leave little room for agency by a new-coming craftsperson. In contrast, leaving identified uncertainties and unexplicit aspects, and favouring a tacit description, would leave room for this agency to be expressed. Learning from carpentry apprenticeship for example, a focus on the gesture and the sensible attention to the material until it *flows in and out*, will favour an uncommoning. The diffusion of the material practice can then be based on principles, like *flour, water, salt, kneading and baking* base methods can be explicit components for bread making or *water, non-iodised salt and chopped down vegetables, fruits, flowers or fungi* to pickling. Preferring principles to fully described recipes is to try and teach based on a tacit to tacit knowledge conversion by companionship. Equivocation is described by Blaser and de la Cadena as a process of *uncommoning* to control because

> commoning often rests on equivocations that disavow uncommonalities, thereby entrenching asymmetries between the elements of the commons. In such situations, uncommoning—patterned after translation as controlled equivocation—might assist commoning in living up to its democratic and equalitarian promise by keeping those involved alert to the uncommons that undergird the commons; calling attention to the asymmetries that translation as *un*controlled equivocation might conceal; creating awareness of the contingent and pragmatic nature of the workability of a translation as controlled equivocation; and committing to mutual transformation (re-description) without having sameness or equivalence as final horizon.
> (emphasis in the original text; Blaser and de la Cadena 2017)

The controlled open-endedness should therefore allow for the commoning. This is a working space that allows a wealth of accidental differences (Eco 2011 p.37) to happen with controlled probes—or explicit knowledge conveyors. It resonates with the ever changing mode of existence of tradition, where pre-individual knowledge meets the expression of the self, thus forming a framework of relational resilience. In the context of the better appropriation of the notion of uncertainty in the students, it can be hypothesised that this equivocation in craft activities facilitated an uncommoning. As the notion of uncommons suggests, the particular accounts regarding uncertainty in the interviews reflect an ontological diversity ranging from a need for scientific description, to a sensible humility to develop towards non-human beings (Rigobello, Gaudillière-Jami and Ayres 2022; appendix F.2). Here, it is worth noting that the student demographics of this program is very international and thus gathers various cosmologies.

Just as designers and architects may find an interest in using mycobosci for their aesthetic qualities, sustainable potential or semiotics, it is important that they are being offered the possibility to engage at multiple levels of knowledge and can evolve through these. The conclusions of this pedagogical study point towards the requirement for further research, such as the monitoring of students appropriation of identified tacit knowledge across a semester with a focus on specific knowledge transmission methods. These methods may be oral, explicit, somatic or *Ba*-oriented (Rigobello, Gaudillière-Jami and Ayres 2022; appendix F.2) for instance, and could focus on the succession of learning phases with regards to knowledge carrier modalities.

4.5 Conclusion

The Anthropocene presents as a state of urgency and motivates to find solutions that can be rapidly applied. But the recourse to the centralisation of technology in the context of an industrial scaling and diffusion of mycobosci displays a risky rationalisation. This reflects directly in contemporary industrial practices, as the identified risks of active chlamydospores global dispersal, development of single substrate and plant monoculture, intrinsic land use potential increase between a mineral or petroleum-based synthetic polymer use, and a plant use, and the energetic use for cultivation are neglected to the benefit of the logistical convenience of the supply chain. Furthermore, the scale of these risks is enhanced by a marketisation presenting mycobosci as sustainable and compostable without conditions, while not supporting these claims by publicly available research and knowledge.

In section 3.5, the plotting of the state-of-the-art reports shows that some of the best performing species, substrate and protocols configurations are not reported. In each of the cases where these materials and methods are not reported in the mycobosci literature, Ecovative is a stakeholder. This exemplifies an effect of lobby mediation, which instrumentalises and shields knowledge to the benefit of commodifying the mycoboscus craft and deprives from the possibility of a public investigation of their activities. The black-boxing of these knowledge also eventually undermines the possibility of a democratic arbitration, should Ecovative's mycobosci production turn ruining for the environment. This is not an accidental output of industrialisation, but the interest for it: to establish a lasting power for capital harnessing. Commodification is integral to the industrial development, because it leverages the naturalisation of a technological infrastructure for a lean scaling of the sales volume. The cost of rapidly implementing an industrial capacity for a product is that of autodetermination capacity loss, dramatic ecological relevance diminishing and political asymmetry. Industrialising in a capitalist framework is to engender ruining ruins both ecologically and epistemologically.

As a way to mitigate the risks presented by the industrial diffusion of mycobosci, a local, opportunistic and energy-saving craft dissemination is recommendable. This is a remarkable opportunity to build upon the greater affordability of the craft (in means and knowledge) and the peer-to-peer social life of fermentation practices. While the review of the colonising effect of Western positivism under global industrialisation is appalling, it had the benefit to trigger the formation of the concept of the uncommons. This is defined as the excess to the commons and that cannot be commoned, i.e. symmetricised for trade. The proposal from Blaser and de la Cadena for the uncommons is useful to identify the breadth and amplitude of what has been reduced to the poor notion of resource in the imperial and economic colonisation processes. To objectivisation, its corollary in subjectivisation is proposed, for the recognition of the principle for an epistemological justice and possibility of a democratic sustainability. Uncommons are qualified as a legitimate intimacy in this study, and identified as integral to social individuation.

The modes of existence of the vernacular designs that were studied revolve around knowledge production and maintenance. These modes support the perpetuation of cosmologies, notably via knowledge transmission methods rooted in craft activities, and the amending of cosmologies by controlled equivocation. It should not be surprising that epistemological autonomy is essential to a community for its ontological sustenance, as vernacular designs live by the condition of affordability in means and knowledge. The knowledge care can be nudged in multiple ways, reflecting on various forms of designed frailties. The introduction of a ritualistic dismantling and rebuilding of a Shinto temple in Shikinen Sengū-sai offers an opportunity to perpetuate a social individuation in a situated religious setting. The re-enactment of the socialised craft in Amish barn raising also shows this quality, but this time does not come with a fixed frequency. It rather comes from the technological limits that the community imposes itself (Bronner 2005). Other forms of fragility may be introduced, and across the examples, they all have in common the perpetuation of a dependence to the social structure. Earlier, a second nature of dependence was identified with geography. But the focus on sustainable institution building that the Gestion de Terroir UN program made contributes to illustrate the primacy of the local and social autonomy over isolated environmental management methods. The program also had a natural resources management aspect as it aimed to fight desertification and impoverishment of the soils from agricultural intensification.

Craftspersonship was identified as central from the review of vernacular designs mode of existence. Allowing for a social individuation, the somatic knowledge transmission and its frequent practice is a modality of controlled equivocation that feeds uncommons. The relational epistemology of craft further contributes to challenge the preconceived idea of tradition as hindering self expression of an individual. It rather grants confidence in the design and bases this on a collectively established situated

knowledge. Tradition displays resilience only in constant change, and one's token contribution to the social identity and practices is thus integral to this adaptation. The capacity of controlled equivocation that lies inherently in craft transmission is key to granting this adaptation and contribution possibility. This conclusion about somatic knowledge is based on the integrative literature review unfolded in this chapter, and served to formulate pedagogical hypotheses about the affordability and transmission of the craft of mycoboscus.
The pedagogical hypotheses were formulated from the explicit and tacit knowledge taxonomy and their conversion modes formulated in a study led by Ikujiro Nonaka, building on an initial conceptualisation by Michael Polanyi. The study was conducted in the context of a master program workshop, and all student groups participated in a survey evaluation of the acquisition of concepts relating to mycobosci. It is hypothesised on the basis of the analysis of the results of the survey and of the student productions that craft activities were more effective for knowledge acquisition than the use of a stochastic simulation model, particularly about the notion of indeterminacy. This illustrates the effect of an expert delegation of knowledge, where the simulation needed a leap of faith from the students with regards to the background scientific knowledge. This study further illustrates the interest for controlled equivocation in pedagogy and the apprenticeship teaching model to avoid an epistemological imposing to students and increase the pedagogical efficacy by leveraging student agency in learning.

This study sets the context for the development of an alternative diffusion of mycobosci to the industrial model. Alternatively to a diffusion of the use of mycoboscus-based products as commodities, a democratic dissemination of the craft is recommended. This study further contributes to bring forth the role of knowledge in social individuation and ontological situating thereof. Based on the findings, contemporary researches about maintenance and its design in architectural contexts could develop effectively about *knowledge maintenance*. Such cautious stirring of the interest, away from a techno-solutionist and objectivist philosophy, towards commoning in controlled equivocation, could contribute to an alternative development and integration of technologies. Such epistemological turn should leverage the political existence of designs, and use knowledge dissemination and retention within local communities to grant agency for a potential democratic evaluation and intervention should they become ruining.

Conclusions & Discussion

In a consistent fashion with the mode of existence of other fermented products, mycobosci originate in logistical needs, and are crafted under a great influence of microbial specifics. If defined early by its capacity to contribute to situated collective intelligence, there is an opportunity for this novel craft to truly become an icon of absolutely sustainable dwelling and design. A mycoboscus description exceeds that of a technology, and by situating culturally the possibility of its sustainable cultivation and use is augmented. The craft can furthermore facilitate the growth of an uncommon excess by complying to vernacular logistic principles.
Across theory and practice, technical and epistemological recommendations have been proposed for this sustainable and cultural potential of mycobosci to advent. The principal contributions, limits and prospects of the studies are presented in the following.

5.1 Contributions

5.1.1 The mycoboscus system

From fungal strain-specific genome to plant material composition, through a wide ranging review as per scales and scientific domains, a comprehensive technical description has been proposed for the mycoboscus system. A first focus on fungi ecological niche, succeeded by a second one on enzymatic activities description, essentially points to the high versatility of the species in use—particularly *G. lucidum*. Inter-individual strain genetic variation and decay performances were also identified, which allowed joint conclusions with that of LCA and risk assessment reviews in drawing a clear recommended manufacturing scenario for mycobosci as local-to-local. From locally sourced byproducts and foraged species, to locally distributed applications—to prevent the accidental spread of chlamydospores from ecologically competitive species—mycobosci add up to a postindustrial contemporary imaginary for design. To the best of the author's knowledge, this is the first time that mycobosci production and use conditions are studied in this breadth and with these conclusions.

Cultivation parameters for mycobosci are numerous and interdependent. For instance, the hydrophilicity of the plant substrate is dependent, in part, on the crystallinity of cellulose for bound water and on wood cell geometry for free liquid water in cavities. Lignocellulosic compounds also soften and dissolve, at different temperatures and in the presence of water, which influences nutrient uptake capacity for fungi. The permeation of fungal enzymes through the wood cell wall is influenced by water activity, and the structure of wood vessels can influence fungal colonisation and decay activities. More parameters have impacts on mycoboscus fermentation, such as nitrogen presence in wood and its migration towards bark upon log drying, for example. The variety of influential variables reflects a potential for fine detailing for mycoboscus design, and a resilience of the system to obtain minimal functional performances. Looking to LCA and environmental risks, if the material system is to be developed as sustainable, then its intrinsic use of plant materials should lead designers and researchers to evaluate carefully the substrates to be used to limit or cancel the eutrophication and acidification potentials, and the levels of other potential soil quality indicators. Resorting only to byproducts of other supply chains can be an impactful leverage in that sense, while the creation of value chain attachments requires the evaluating of ripple effects and should be aware of the economic dependencies that it might sustain. The opportunistic use of materials is recommended in that sense.

The systematic review of the mycobosci state-of-the-art, along with the semi-systematic review in material science and fungal biology, resulted in the identification of three design strategies for mycobosci: supplementation, densification and composition. These strategies can be used to structure a system around the local-to-local model and reduce the elemental risk in substrate variability by otherwise increasing the mechanical performances of composites. Developing common knowledge and material design principles via the supplementation strategy can help this, that is from an opportunistic view—supplementing to adapt a protocol to local material sourcing. Densifying mycobosci is an appreciated strategy in the state-of-the-art as it is an effective method to increase strength and stiffness. Although this specific compressing process has not been evaluated yet through LCA, studies in uncompressed mycobosci show a high sensitivity of the system to electric energy use (particularly at sterilisation, incubation and drying stages) and suggest that to produce a sustainable composite that is at least neutral in CO_2 eq. emissions, passive manufacturing processes need to be developed. This is also valid for densification processes. Composition strategies then cover aggregate design, positioning, and macro-structuring. The arming of composites can be achieved with significant effect, and provides a more efficient manufacturing method compared to densification as per embedded energy.

5.1.2 Composition strategies

As fungal species and substrate materials should be sourced and distributed locally to be sustainable and prevent environmental risks, this will cause inconsistencies and reproducibility challenges. These practical mechanical frailties can be addressed by implementing mycobosci composition strategies. These strategies, scarcely studied yet, are presented and critically discussed as ways to modify and control the mechanical behaviour of mycobosci composites. A novel approach, introduced in this thesis, is the stiffening effect produced by the incorporation of arming large fibres. This has been shown to lead to outstanding bending stiffness results in comparison to the state-of-the-art. The rattan material that was used in the studies should be replaced by a locally-produced and upcycled material beyond laboratory conditions. This experimental design (BM_R) is still approximately three times more elastic and 16 times weaker than MDF, although it is approximately three times less dense. The introduction of rattan short fibres as failure initiators also proved to have a significant effect for modifying the compressive stress response. Hessian jacketing was tested in compression and tension; although it had a negative impact on compressive stiffness and strength, it contributed to increase bending stiffness and strength in the medium-sized particles group (BM). This jacketing can be an effective method to introduce local tensile reinforcements or contain the infill of a composite that might break apart, and is hypothesised to be contributing to the durability of mycobosci. The specific bending stiffness of the rattan-reinforced group

and hessian-jacketed group are situated above the considered state-of-the-art (at respectively 5.37 and 1.59 $10^3 m^2 s^{-2}$), thus contributing to demonstrate more efficient mechanical designs with regards to the load case. The departure of these data from the state-of-the-art pool is illustrated in Fig.3.48. The contribution of these mycoboscus designs to the expanding of a new material research area made of the upper merging of *natural materials* properties and *foams* properties is illustrated in Fig.3.46. The introduction of macro structures in the experimental specimens has also resulted in the modifying of composite dimensional stability during drying. This feature can find usefulness in the future for production control and creative uses. Aggregate size effect was tested, and the smallest granulation that was used resulted in significantly lower performances (0.5 – 1.0 mm). The best performing aggregate size (0.75 – 3.0 mm) is also the one having the highest maximum particles volume fraction (supplier data: table 3.15, specimens-based data: table 3.16).
Building upon the rule of mixture for determining two-phase particulate composites elasticity, a method for analytical determination of the specific matrix modulus has been proposed. Based on experimental quantification of tensile stress material response, this metric provides information on the quality of the binding resulting from the cultivation. In this work, it was useful in analysing aggregate size effect on the fermentation quality, and to conclude that medium and large aggregate classes resulted in a same binding quality as per stiffness. This is the first report of such method for mycobosci to the best of the author's knowledge. In the perspective of a wider adoption of the mycoboscus craft by amateur practitioners, this frugal determination method will be useful to develop quick and low-cost design and evaluation methods.

5.1.3 Design terroir

Mycobosci have been the object of commercial applications since 2007, as patented alternatives to polystyrene solutions for example. Conceived in a green growth framework, this resort to industrial development is based on the protection of an industrial property and withholding of knowledge. The highly affordable mycoboscus technique sees its transformative and sustainable potential cut short by being commodified. Furthermore, the expert mediation that an industrial development enacts impedes the capacity of public scrutinising and democratic intervention, should industrial mycoboscus production turn too polluting or socially detrimental. This is a present situation with the monopolistic industrial actor which utilises mycobosci now, as its designs do not mitigate but increase the ecological risks that the technology presents.

With a focus on knowledge production and transmission, vernacular design modes of existence have been reviewed to construct an alternative use diffusion strategy to industrialisation. In this strategy, the risks mitigation necessitate the support of the development of local economies, and opportunistic and energy-saving designs. The concept of the bioregion is a recommendable framework for developing these.

This thesis coins the expression design terroir. The concept elaboration is build upon the millenia-old eco-social design lineage, and the ancient knowledge about the cultural geography of terroir. Of course, the affordable fermentation system of mycoboscus echoes in the crafts of bread making and cheese production, which productively influenced this conceptualisation.

A design terroir is bioregionalist, fragile and based on craftspersonship. It leverages a cosmological situating, and a model for authorship and ownership that is relational. The frailties that are present in diverse designs have been analysed from the perspective of the opportunities they grant for care practices. The idea of design terroir was shown to particu-

larly leverage a relational epistemology, and the maintenance of frailties are generative of an uncommon grounding in somatic tacit knowledge. The modernist conception of tradition as conservative is particularly challenged in this epistemological view, for as the individual inherits from a pre-individual knowledge, the apprenticeship model of transmission is majoritarily based on leveraging change in controlled equivocation.
Alfred Gell comments on the internalisation of prow board carving know-how in the Trobriand society (Papua New Guinea). A magical ritual aims at educating the craftsperson into not only being skilful in shaping the matter with primitive tools, but overcoming an inherent physical resistance such that they can project an aesthetic that is commissioned by the community. The person is expected to add a token contribution to a series. To get trained into this practice, they are educated through a spiritual procedure aiming at having the forms to inscribe *flow in and out* of their gesture (Ingold 2001). After having experienced other water and liquid-anchored rituals, they finally drink snake blood, to "*grow into sinuous from within*" (Gell 1994). The enchantment of technology, as designated by Gell, regards not really the craft outcome, but the *mode of production as mode of individuation*. In light of the present study, this enchantment is analysed as an ontological contribution to, and care of, situated uncommons. The socially anchored crafts practice therefore fundamentally contributes to a social individuation in this context.

The findings from the review works in chapter IV led to the formulation of epistemological hypotheses that were experimented in an architectural education context. The results of this study indicate a better acquisition of tacit knowledge about mycobosci through craft activities than through the use of a simulation model. The results contribute to display the leap of faith effect that expert mediated teaching supports favour for a generalist audience, in the context of the simulation model use. This leap is a naturalisation, and was adversarial to the satisfaction of pedagogical goals. In section 4.2, this naturalisation effect was identified to be central to the commodification of knowledge integral to industrial development.

The interest for this experiment was twofold. Its primary interest was pedagogical; it was motivated by gaining direct experience about the effect of craft tacit knowledge transmission in a controlled equivocation model—i.e. an apprenticeship model. This pedagogical area of research can be developed towards evaluating other modalities such as oral, explicit, somatic or *Ba*-oriented (Rigobello, Gaudillière-Jami and Ayres 2022). The second interest concerned the capacity of these activities to support a relational epistemology and contribute to support alternative ontologies to modernist and cybernetic ones. This is to oppose a humble, sensible eco-social resilience development model to the objectivist colonialism and technological solutionism that characterises these philosophies.

5.1.4 Design research

The mix of methodologies employed in this thesis illustrates an anchorage of design research both in technique and its cultural effects. The novelty of mycobosci offers a rare context of study for a design medium, much like the discovery of a novel land to understand. The economic and semiotic entanglements for the mycoboscus domain are scarce compared to the craft of carpentry for instance, besides the presence of a few established commercial actors. And from this novel land, a perspective over the previously mapped domains can be drawn. The object of mycoboscus was considered as a vantage point to undertake the mode of existence of design, both generally and with interest for this singularity.

The overall integrative approach resembles a prescriptive anthropology of technique. The use of an integrative review method supported an iterative sense-building and situating during the research process, and can further allow a systematic approach to form a design sense for a given research question. The synthesis that the method facilitates supports the integration of findings heterogeneous in their nature, under a condition of symmetry between areas—articulated by the research question. This condition is a hypothesis formulated on the basis of the critical role that the review of an environmental risks assessment and LCA studies played in the funnelling of recommendable scenarios for mycobosci democratisation, which echoed in the findings made from economic and anthropological reports. The matrix approach reflects the systemic view that *design* invites to, and allows for in through its characteristic synthesis activities. The structuring of this work contributes to design research methodology by illustrating the converging use of heterogeneous methods around a technical topic in a praxeological manner.

5.2 Limitations

Although the vast majority of the mycobosci mechanical property effects studied experimentally were statistically significant, the improvement of manufacturing conditions for mycobosci is expected to lead to a reduction in standard deviation of future results. Nonetheless, efforts were already made to facilitate reproducibility and increase analytical readability through the use of geometrically regular materials in the production of specimens. As the findings point to the recommendation of addressing an increased complexity in raw materials for future investigations, through byproduct upcycling for example, developing a comprehensive and systematised approach to the chemical and mechanical qualifying of aggregates will be useful. Moreover, as conducted in chapters II & III, the frequent research into the evidence-based practices of the field of mycobosci will be critical to inform the definition of use-case attractors for material specification and to support the informed advancement of research with metascience. In section 3.4.3, the significant difference between the BM and HC specimen groups and the opposite effect that reinforcements had in the results from the HC group (besides rattan arming) highlight the need for a better availability of knowledge regarding production controls. The parametricisation of manufacturing protocols can help researchers in the future to identify statistically important parameters so as to efficiently improve quality—for instance, using Taguchi methods (Rao *et al.* 2008).

Some of the raw materials that were used in the design of specimens include rattan, a species that is not native to Europe. The choice of substrates in the present work was made for reducing experimental variables and increasing reproducibility, occasionally at the cost of being sourced globally. In the future, an LCA-informed scoping of raw materials for mycoboscus studies should be systematised to support sustainable designs early. The experimental production methods involved sterilisation, incubation and oven-drying which were reported in the literature to make mycobosci unsustainable. While the scientific review in chapter II reveals the number of influential variables on the manufacturing process and the need to reduce those by ensuring a level of production control, designing future studies under strict conditions of sustainability seems important.

The macro composition strategies that were tested in chapter III only cover a few designs, some of which did not prove significant. This is the result of a decision not to focus on mechanical optimisation but to probe different typologies. Similarly, only particles and not fibres were considered as principal substrate. Considering the positive contribution of fibres to tensile stress response, future mechanical optimisation studies may qualify these further with byproducts use.

The research presented here was conducted almost exclusively with a single strain of the species *G. lucidum*. The choice was made to produce results that are coherent between experimental series and as a means to deepen the somatic tacit knowledge rather than widen it. While this is a form of limitation, it was also a methodological choice backed by the fact that this species has the largest known CAZymes array.

The lessons from the Gestion de Terroir efforts show how political influence can undermine systemic enterprises aimed at implementing sustainability or regenerative solutions when a delegation of power is implied. The study of global inequalities led by Dorninger also presents overwhelming evidence of complex logistic and economic entanglements that make humanist efforts for eco-social development look modest—which may motivate those as a result. The local-to-local technical recommendation found an echo in vernacular designs modes of existence, and other models of supply chain definition can be comparatively analysed in greater detail in the future with regards to environmental risks, aesthetics, design epistemology, ontological situating and means of risk mitigation. The notions of authority and control in the modes of existence of technologies and knowledge maintenance were mentioned but not fully treated. For example, the effects of a social individuation grounding on somatic tacit knowledge and an excess of sensible qualities on discourse legitimacy in the social sphere, ecological situating and social inclusion, were studied but not elaborated upon in this manuscript (Rigobello and Gaudillière-Jami 2021). The epistemological consequences should be further investigated in future researches in the areas of aesthetics, anthropology, sociology and philosophy for advancing design studies.

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5.3 Research directions

It was stated that mycobosci illustrate a simple and affordable technique. This means that the fermentation principles are versatile in relation to the varied levels of equipment and material sourcing that craftspersons may have access to. Mycobosci resemble the craft of cheese in this regard, and their technical and aesthetic range has yet to be uncovered. For mycobosci to have a chance at practically and culturally contributing to eco-social development, the dissemination of the craft needs to comprise comprehensive fungal biology principles. Future studies on the modality of the craft adoption and the expansion of locally-informed uses should support an informed democratisation. Because of the novelty of this craft, the development of a *prescriptive reversed anthropology of technique* angle for design studies will find a wealth of information in the study of the dynamics of a progressive technical adoption. Such study of local mycoboscus craft and use dynamics will represent a unique field for STS researchers. Future platform tools designed to facilitate collective knowledge augmentation and community autonomy will be useful for this.

Technically, the studies of the inactivation conditions for chlamydospores of all species in use for mycoboscus production are a priority as environmentally competitive species are used and spread globally now. The compostability of these composites is actively promoted without caution at the moment. Moreover, and following the findings from LCA studies, the development of energetically passive processes for cultivating mycobosci should be invested in as a means to make them unambiguously CO_2 eq. neutral or negative, and lower their other environmental impacts such as the acidification potential. It can be imagined that the open documentation of such thrifty processes would further augment the adoption rate of the technique in communities.
The knowledge about the supplements that have been identified for regulating fungi enzymatic activities will benefit from a systemic experimental study to determine their effect on mechanical performances such as binding strength. This should result in local recipe optima per species and principal substrate couplings. But the knowledge that these supplementation studies are expected to generate should also benefit the

creative design of artefact aesthetics to be ripened, and as a means to modify the synthesis of volatile organic compounds in mycobosci.

Specific cultivation protocols may be developed in the future using knowledge of enzymatic activities, by instrumentalising the capacity of fungal selective delignifiers to primarily degrade lignin for instance. To this end, ligninase activities can be increased by using syringic acid (LiP, laccase), gallic acid (MnP), H_2O_2, $CuSO_4$ (laccase), $MnSO_4$, or surfactant to facilitate enzyme permeation and the solubilisation of lignin. Although fungi cannot feed from lignin, this development could focus on keeping crystalline cellulose integral to engineer the composite structure and mechanical behaviour.

Because this is a knowledge gap in mycobosci literature, the future experimental qualification of the decay of lignin and hemicellulose in plant substrates will be useful to refine mechanical models. Lignin is a primary contributor of strength parallel to grain and hemicellulose of compressive strength perpendicular to grain; their decay affects aggregate hardness.

The specific matrix modulus metric, introduced in this thesis, can be useful to investigate the effect of aggregate size and shape over the fermentation activity and resulting binding quality efficiently and in greater detail. It could also be used in future studies to evaluate the effect of supplements on grown mycelia. Reporting on this metric necessitates little analytical effort. Its use could therefore be democratised so as to increase detailed knowledge about the craft over time.

Hybridising between the three strategies of supplementation, densification and composition will lead to outstanding mechanical performances for mycobosci in the future, making them more reliable material solutions. The democratisation of macro composed designs in recent years has already resulted in a variety of complex designs contributing to raise interest for this track (Nguyen *et al.* 2022; Özdemir *et al.* 2022). More creative layouts will surely be demonstrated and percolate to market solutions, while researching product design that hybridises with woods, ceramics and metals should see an interest for advancing interaction design given the unique haptic of mycobosci. An example utilising upcycled materials is depicted in Fig.9. At the moment, these materially hybrid strategies have only scarcely been explored. Describing the modalities of durable assemblies of mycobosci and other materials will be critical for their market-readiness.

The contemporary adoption success of industrial technological solutions pairs with a growing socio-technical entanglement. Moving forward, the study of the conditions for the subsistence and disruption of these logistical links by way of design and engineering should help to identify effective actuators to support their democratic rethinking (Bonnet *et al.* 2021). On-going discussions on open design & engineering and active citizen participation already illustrate promising methodologies echoing in the mode of existence of traditional designs (Hui and Halpin 2013). A continued research investment on the epistemology of these practices and their effectiveness for social individuation and community autonomisation will be useful to advance social justice and the sustainability of our fabrication and use practices at once.
While an understanding of the relational nature of technique has been slowly infusing in design theory—with a social justice perspective as found recently in Escobar (Escobar 2018)—it has yet to become a dominant interest and model in design activities. The growing popularity of systemic design should find a fertile ground in developing practices that resort to relational aesthetics for eco-social sustainability and resilience. For this, the epistemological interest in standardisation and individuated experience can be explored further in research works.
A relevant strand of research for designers, can be to consider in greater detail the reduction of inequalities in a world of restricted economies. Recent works question the possibility of ensuring social minima against

environmental ceilings, while questioning the global living standards that should be aimed for in an equitable world (Chakrabarty 2021). Useful for this project, historian Dipesh Chakrabarty further develops a political history of non-Western driven modernisation in India, which contributed to raise living standards and develop India's global geopolitical position (Chakrabarty 2021). The possibility of a non-Western modernisation of medical practices has been the object of a recent study in the Indian state of Kerala. Indeed, the governments of this state have been developing primary health care (PHC) practices in advance of the world health organisation (WHO) recommendations, and building on traditional medical and pharmaceutical knowledge along with recent scientific knowledge (Beaudevin *et al.* 2023). Exploring the modalities under which design science and practice could learn from and support technological and epistemological autonomy for reducing inequalities while developing sound ecological policies, is an enticing project. As was illustrated in this manuscript, the role of design activities in redirecting Western living standards to a sustainable level can be ported to the area of epistemology and ontology in design with a political project:

> Then, like a flag raised on a pole, the standard offers nothing more than a modest salience around which to rally.
> (Thévenot 2009)

5.4 List of academic dissemination

5.4.1 Peer-reviewed journal articles

Compressive Behaviour of Anisotropic Mycelium-Based Composites.
A. Rigobello, P. Ayres.
Scientific Reports, April 2022.
DOI: 10.1038/s41598-022-10930-5
Appendix D.1.

Effect of Composition Strategies on Mycelium-Based Composites Flexural Behaviour.
A. Rigobello, C. Colmo, P. Ayres.
Biomimetics, April 2022.
DOI: 10.3390/biomimetics7020053
Appendix D.2.

5.4.2 Peer-reviewed book section

Design Strategies for Mycelium-Based Composites.
A. Rigobello, P. Ayres
Fungi and Fungal Products in Human Welfare and Biotechnology,
Springer Nature.
DOI: 10.1007/978-981-19-8853-0_20
Appendix E.1.

5.4.3 Peer-reviewed conference articles

Fragile Computation: Rethinking Information Technologies to Foster Situated Ecologies.
A. Rigobello, P. Ayres.
Deep City Conference Proceedings, March 2021.
In-press.
Appendix F.1.

***Prototaxites stellaviatori*: a Fungal Growth Simulation Model for Mycelium-Based Composites Education in Applied Arts**.
A. Rigobello, N. Gaudillière-Jami, P. Ayres.
Structures and Architecture (ICSA 2022), July 2022.
DOI: 10.1201/9781003023555-5
Appendix F.2.

Designing the Gross. In Search for Social Inclusion.
A. Rigobello, N. Gaudillière-Jami.
Design Culture(s): Cumulus Conference Proceedings Roma 2021, April 2021.
ISBN: 978-952-64-9004-5
Appendix F.3.

5.4.4 EU H2020 report

D5.2 Report on preliminary characterisation and modelling.
Lead author.
November 2021, Fungal Architectures (Grant agreement No. 858132).

5.4.5 Lectures and symposium interventions

Interspecies Cosmotechnics, September 2021.
Mycotecture Talks, Newcastle University (UK).
(Ozkan 2021)

Designing the Gross, August 2021.
Mycelium Mini-Meeting, RISE (SE).
(Attias *et al.* 2021)

The Fungal Momentum, August 2021.
Summer Sessions: Empirical Spirits, Little Red Door (FR).

Fungal Hygge, May 2021.
Spaces of Welfare International Conference, Royal Danish Academy (DK).

A Rhythm for the Commons, April 2021.
Rhythm: Affect, Technology, Aesthetics Seminar, SDU (DK).

Fungal Materials in Inclusion, December 2020.
Fab City Nancy Studio, ARTEM Nancy (FR).

Models as Tactical Tools, November 2020.
Louvre Writter Studio, TU Darmstadt (DE).

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Appendices

A Illustration

A.1 Annotated multi-scalar mycoboscus model

A3 poster attached to the manuscript.

B Technical datasheets

B.1 Räuchergold type HB 500-1000

Typenbeschreibung

Type
HB 500-1000

RÄUCHERGOLD®

natürliche Holzfaserstoffe

Ausgangsrohstoff

ausgesuchtes Laubholz

Physikalische und chemische Eigenschaften

Farbe		hellbraun
Struktur		faserig
Partikelbereich, Hauptteil		0,5 - 1,0 mm
Schüttgewicht		180 g/l - 250 g/l
Glührückstand (850 °C, 4 h)	ca.	1 %
pH-Wert		5,5 +/- 1

Siebanalyse

Siebrückstand (mit Engelsmann - Rüttelsieb) bei einer lichten Maschenweite von:

1,25 mm	0,63 mm	0,5 mm
max. 5 %	max. 65 %	max. 90 %

Wie bei allen natürlichen Produkten können sich geringfügige Abweichungen von den oben aufgeführten Werten ergeben.

Allgemeine Hinweise

RÄUCHERGOLD® -Holzfaserstoffe sind umweltfreundliche Produkte, die aus nachwachsenden Rohstoffen gewonnen werden.

J. RETTENMAIER & SÖHNE GMBH + CO
Fasern aus der Natur
Holzmühle 1
D-73494 Rosenberg

Telefon: **0 79 67/1 52-0**
Telefax: **0 79 67/1 52-222**

B.2 Räuchergold type HBK 750-2000

Typenbeschreibung

Type
HBK 750-2000

RÄUCHERGOLD®

natürliche Holzfaserstoffe

Ausgangsrohstoff

ausgesuchtes Laubholz

Physikalische und chemische Eigenschaften

Farbe		hellbraun
Struktur		kubisch
Partikelbereich, Hauptteil		1,2 - 1,8 mm
Schüttgewicht		270 g/l - 330 g/l
Glührückstand (850 °C, 4 h)	ca.	1 %
pH-Wert		5,5 +/- 1

Siebanalyse

Siebrückstand (mit Engelsmann - Rüttelsieb) bei einer lichten Maschenweite von:

2,5 mm	1,6 mm	1,0 mm
max. 5 %	max. 55 %	max. 95 %

Wie bei allen natürlichen Produkten können sich geringfügige Abweichungen von den oben aufgeführten Werten ergeben.

Allgemeine Hinweise

RÄUCHERGOLD®-Holzfaserstoffe sind umweltfreundliche Produkte, die aus nachwachsenden Rohstoffen gewonnen werden.

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B.3 Räuchergold type KL 2-16

Typenbeschreibung

Type
KL 2-16

RÄUCHERGOLD

Natürliche Holzfaserstoffe

Ausgangsrohstoff

Ausgesuchtes Laubholz

Chemische und physikalische Eigenschaften

Farbe		hellbraun
Struktur		kubisch
Durchschnittl. Partikelgröße / Partikelbereich		6 mm - 10 mm
Oxidasche (850 °C, 4 h)	~	1 %
pH-Wert (10 %-ige Suspension)		4,5 - 7
Schüttgewicht (in Anlehnung an DIN EN ISO 60)		220 g/l - 320 g/l

Siebanalyse (mit RETSCH-Vibrationssieb) bei einer lichten Maschenweite von:

> 15 mm	> 8 mm	> 5 mm
0 %	max. 20 %	45 % - 85 %

Allgemeine Hinweise

RÄUCHERGOLD Holzfaserstoffe sind umweltfreundliche Produkte, die aus nachwachsenden Rohstoffen gewonnen werden.

Wie bei allen natürlichen Produkten können sich geringfügige Abweichungen von den oben aufgeführten Werten ergeben.

Ausgabedatum: 2013-07 / 1 Seite

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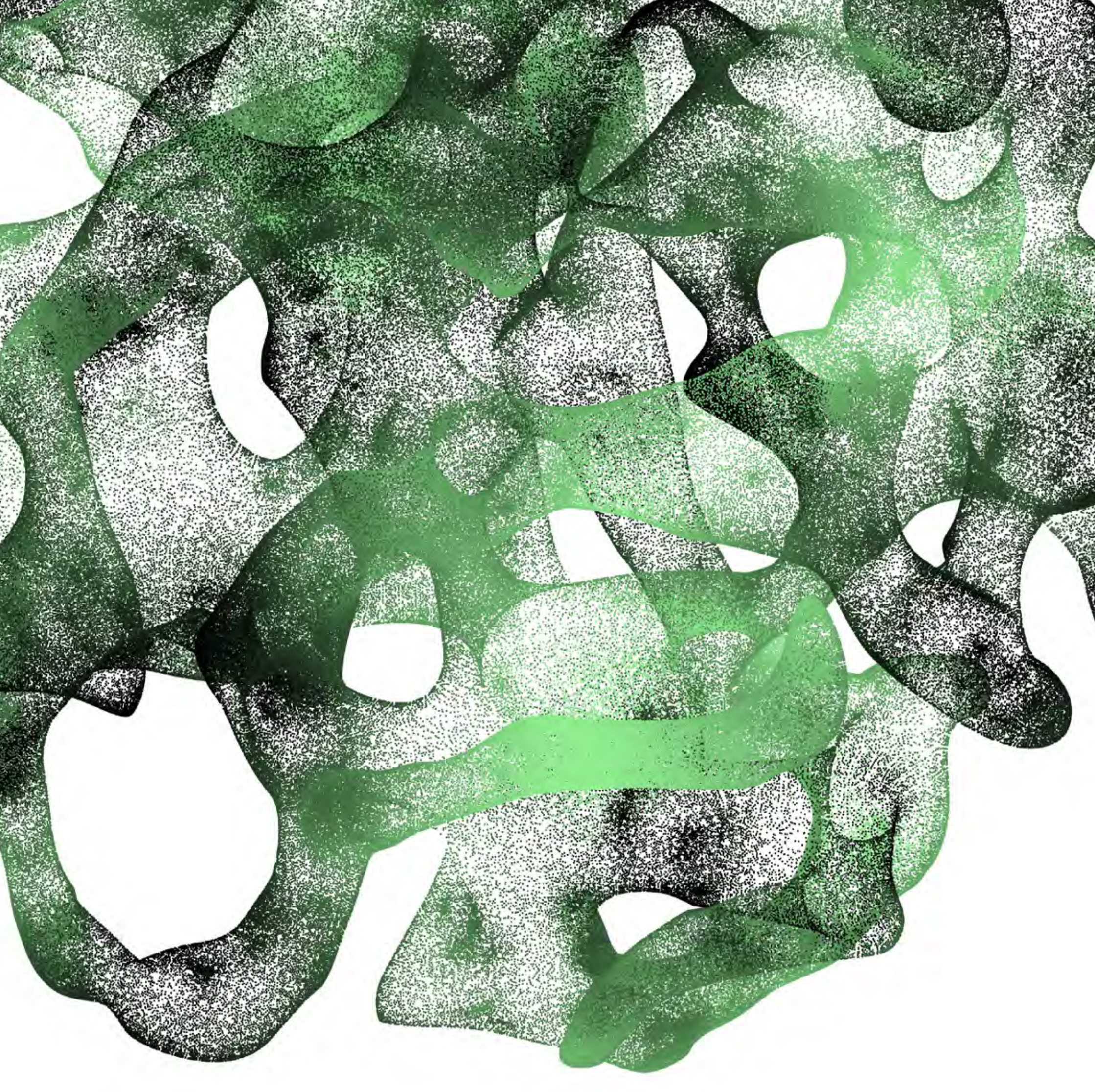
C Pedagogical material

C.1 Liquid Modelling students guide



liquid modelling,

navigation beyond vision.

Computation in Architecture 2020

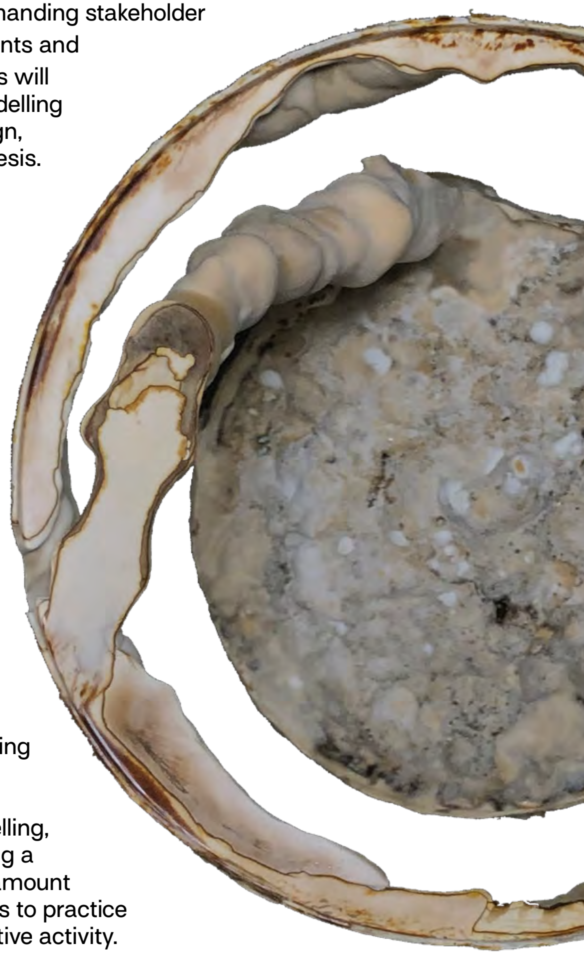
Fungal Architectures

CITA

Document author: Adrien Rigobello (CITA, KADK).
Issue date: September 17th, 2020.

Fungal Architectures is a research project which aims at implementing a fungal automaton at the architectural scale. From the speculative context that it offers us, students will be introduced throughout this workshop to **stochastic modelling** workflow to support **mycelium-based designs**. After a first workshop through which students will build a sensitivity to the craft of mycelium composites making, this one will focus on predicting mycelial outcomes from complex substrates compositions.

Beyond the necessary technical sustainability of architectural projects, it is the porosity of designs to other living beings with differing environmental requirements that we will investigate. This hands-on workshop will be the opportunity to question the role that agency delegation through **indeterminacy** embedding could play in the act of architectural synthesis: how much design agency do you desire to confer a mycelium? For it just became a demanding stakeholder in your design. Between hands-on experiments and computational simulation, students as groups will develop critical thinking on phenomenon modelling as a prevalent activity in computational design, and on agency delegation in the act of synthesis. Through making, students will further develop a sensitivity to living materials – ones that embody propositions for post-anthropocentric designs.



The main parameter to map for predicting mycelial growth is the **moisture** within the substrate. But even though we can attempt to predict the mycelial outcomes, this modelling is an activity that challenges our knowledge about each part composing the system (fungal strain behaviour, environmental modelling, composites modelling, to name a few). Facing a tremendous modelling complexity with this amount of variables is a chance to challenge our ways to practice modelling in architectural design – as a creative activity.
The context of **Fungal Architectures** necessitates to work with living fungi in a work of architecture offers a space for discussing post-anthropocentric designs: how to offer a shelter for fungi to foster, as much as for humans to do so? The scientific and architectural paths meet around this notion of uncertainty, of probabilities, of questions rather than affirmations. We suggest an elusive modelling approach to the problem of predicting fungal growth, using stochastic modelling.

Cover: stochastic simulation of mycelium growth (A. Rigobello, FUNGAR, 2020).

Fruiting body of fungus *Ganoderma resinaceum* emerging from a mycelium composite experiment (A. Rigobello, FUNGAR, 2020).

agenda

	M	T	W	T	F	S	S
w 38	14.09	15.09	16.09	17.09	**18.09**	19.09	20.09
Stage					DESIGN	DESIGN	DESIGN
09:30					Models as tactical tools *Lecture*		
10:00							
10:30							
11:00					Fungal prediction simulation intro *Tutorial*		
11:30							
12:00					Integrating models into the simulation *Hands-on*		
12:30							
13:00							
13:30					Studio meeting		
14:00							
14:30					Simulation model fine tuning *Tutorial*		
15:00							
15:30					Fine tuning of models within the simulation *Hands-on*		
16:00							
16:30							
17:00							
17:30							

	M	T	W	T	F	S	S
w 39	21.09	22.09	**23.09**	**24.09**	**25.09**	26.09	27.09
Stage	DESIGN	DESIGN	DESIGN	FAB	SENSING	SENSING	SENSING
09:30	Written Assignment (5th year)	Written Assignment (5th year) Rhino/GH Fundamentals (4th year)	Groups project quick review (5min per group)	Project work *Hands-on*	Project work *Hands-on*		
10:00							
10:30			Project work *Hands-on*				
11:00							
11:30							
12:00							
12:30							
13:00							
13:30							
14:00	Written Assignment (5th year)	Written Assignment (5th year)	Project work *Hands-on*	Project work *Hands-on*	Project work *Hands-on*		
14:30							
15:00							
15:30							
16:00							
16:30							
17:00				Groups project quick review (5min per group)			
17:30							

	M	T	W	T	F	S	S
w 40	**28.09**	**29.09**	**30.09**	01.10	02.10	03.10	04.10
Stage	SENSING	CONCLUSIONS					
09:30	Groups project quick review (5min per group)	Project work *Hands-on*	CRIT	Intro GH (4th year)	Intro GH (4th year)		
10:00							
10:30	Project work *Hands-on*						
11:00							
11:30							
12:00							
12:30							
13:00							
13:30							
14:00	Project work *Hands-on*	Project work *Hands-on*	CRIT	Intro GH (4th year)	Intro GH (4th year)		
14:30							
15:00							
15:30							
16:00							
16:30							
17:00							
17:30							

organization

Horaries:
Each teaching day will commence at 09h30 sharp. Please ensure you have arrived and are settled before this time. Morning sessions will run until 13h00 to coordinate with the rules related to the canteen. Access to the canteen is restricted and staged by year of study. This means that year 4 students will have access from 13h00-13h30. And year 5 students have access from 13h30-14h00. The afternoon teaching sessions will commence at 14h00 sharp. In non-teaching periods you are expected to continue responding to the brief and pushing forward with your investigations. Remember that access to the studio and biolab is strictly limited to 08h00 - 20h00 weekdays.

Location: CiA studio, Biolab.
Workshop leaders: Adrien Rigobello, Phil Ayres, Claudia Colmo, You-Wen Ji.

system config

Please install ahead of the workshop the following Grasshopper plugins, and the latest Windows update of Rhino 6:

- **Anemone**; https://www.food4rhino.com/app/anemone
- **WeaverBird**; http://www.giuliopiacentino.com/weaverbird
- **Dendro**; https://www.food4rhino.com/app/dendro
- **Volvox**; https://www.food4rhino.com/app/volvox
- **Topologizer**; https://www.grasshopper3d.com/profiles/blogs/topologizer-network-cleanup
- **P. stellaviatori;** (download link)

How to install a Grasshopper plugin: https://www.giancadm.com/plugins-install

outcomes

Required group outcomes:
DESIGN
- Modelling hypothesis (R, G, B and their probabilities),
- Key experimental metrics to monitor,
- Testing protocol,
- Documentation of design iterations,
- Description of the Moisture and Fibres setup,
- Location choice, and solar analysis results (1 to 3 volumetric point clouds),
- Formal enquiries on uncertainty.

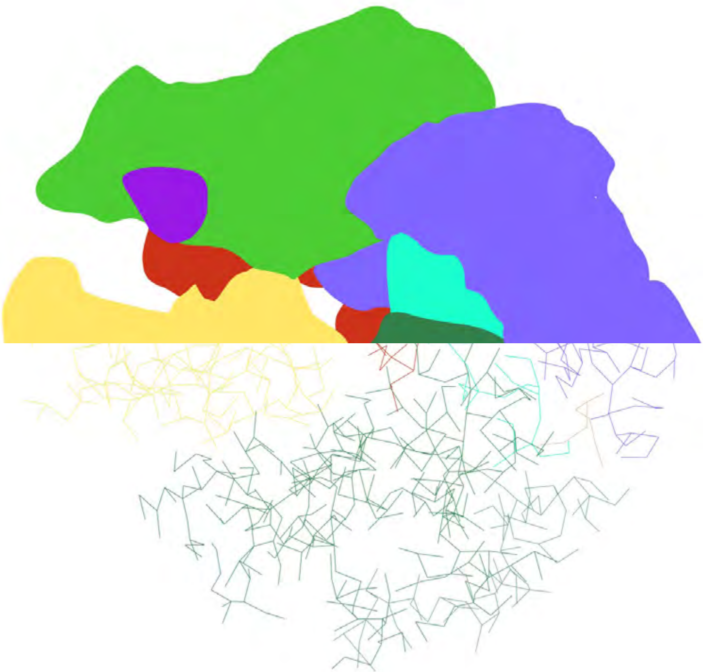
FABRICATION
- Fabrication protocol for the fungal artefact,
- Actual artefact.

SENSING
- Sensing protocol,
- Sensing dataset, and observations.

CONCLUSIONS
- Conclusions on experimental observations against simulation results,
- General discussion on the craft (ex.: have you smelled a contamination?).

Documentation resources should take the form of:
- Group log book on Google doc,
- Final workshop catalogue (packaged InDesign file),
- Powerpoint presentation.

Learning objectives:
- To gain competence in assimilating existing research insight and utilising it in a design context.
- Competence in critically reflecting upon architectural issues through direct material engagement.
- Competence in developing appropriate representational methods and tools.
- Competence in deploying appropriate fabrication strategies for addressing material and fabrication-based issues.
- Use of appropriate fabrication technologies to support design investigation and synthesis.
- Knowledge of methods for testing and evaluation of material performance
- Knowledge of simulation techniques, their assumptions, abstractions, limits and opportunities within design contexts.
- Skill in applying core concepts of digital practice related to design, analysis, simulation and fabrication.
- Competence in working effectively within a group-working context.



what are fungi?

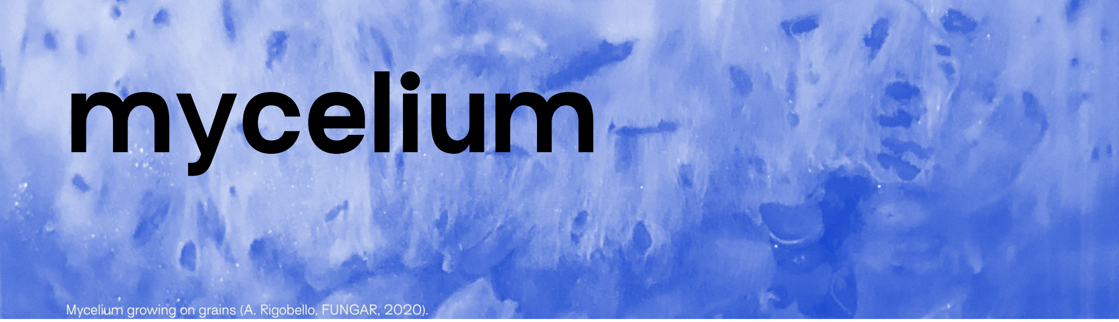
The Kew Royal Botanical Gardens (UK) is editing a yearly report called «State of the World's Fungi»; here is an exerpt from the 2018 edition:

« Fungi are distinctive organisms that digest their food externally by secreting enzymes into the environment and absorbing the dissolved organic matter back into their cells. Most have cell walls composed primarily of chitin (a substance that is also found in the animal kingdom, for example in the exoskeletons of insects and shells of crabs and lobsters). They also store food reserves as glycogen and lipids (not starch as in plants). Thus, despite the superficial resemblance of some fungi to plants (e.g. having rooted, stalked structures), their non-photosynthetic, absorptive method of feeding and their different cell walls, cell membrane chemistry, methods of food storage and DNA indicate that they form an independent [biological] kingdom.
Some fungi exist as microscopic, single-celled yeasts (e.g. the bloom on the skin of a plum or grape), while the most complex forms have a far more elaborate multicellular body comprising an interconnected network, or **mycelium**, of minute, protoplasm-filled tubes called hyphae. The individual thread-like tubes extend at their tips and form branches that explore their environment, fight with other fungi to occupy territory, or interact with other organisms. These activities can occur inside a few cells of a leaf, in a column of decay extending for several metres inside a tree trunk, or in the soil, for example forming a giant 'fairy ring' of mushrooms in ancient grassland.
Other fungi live as lichens – a symbiotic association between a fungus (the mycobiont) and at least one photosynthetic partner (the photobiont), which can be an alga, a cyanobacterium or both. Lichens are often referred to as the ultimate example of mutualism (a type of symbiosis in which both partners benefit), given their ability to form distinct biological entities so well integrated that they resemble one single organism. They grow almost everywhere; they can be found in most terrestrial habitats, even in extreme conditions such as Antarctic deserts, growing on rocks, bark, soil, leaves, mosses, man-made materials and even on top of other lichens.

[...] Fungi are associated with the roots of almost all plants, including forest trees and most food crops – the fungi act as living intermediaries between the plant and the surrounding soil. This type of root–fungal interaction is known as a mycorrhiza and, like lichens, the partners engage in a mutualistic relationship. The plant benefits from the greater capacity of the fungus to absorb water and nutrients and to mobilise minerals that would otherwise be unavailable, and the fungus benefits from a steady source of carbohydrates from the plant.
Fungi are also the most significant organisms that break down **cellulose**, **hemicellulose** and **lignin**. These are the tough polymers in plant cell walls that give wood its great strength and durability. Their decomposition by **wood-decaying fungi** releases key plant nutrients back into the soil, thereby allowing the next generation of seedlings to grow. Without nutrient cycling, life on Earth as we know it would not exist; nutrients would be in such short supply that biological growth would be severely limited right across the globe. »

Fungus from the family *Psathyrellaceae* decomposing bark chippings (J. Eden, Royal Botanic Gardens Kew, 2018).

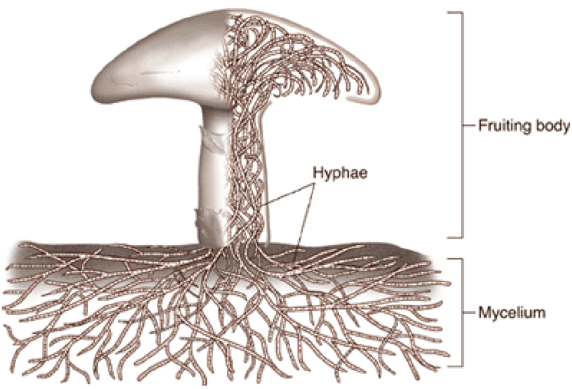
mycelium



Mycelium growing on grains (A. Rigobello, FUNGAR, 2020).

Mycelium is composed of a network of hyphae, which can essentially be thought of as the vascular system of the fungi. In *Ganoderma* species, their diameter ranges from 5 to 7.5 micrometers (at least three times smaller than a human hair). They can be observed with a naked eye (see the picture above as an example, where we see the white hyphae «hairs» composing the mycelium colonizing the substrate from top to bottom). Mushrooms, which are fruiting bodies of certain species of mycelium (yeasts, which are fungi, do not produce mushrooms for example), are also composed of mycelium.

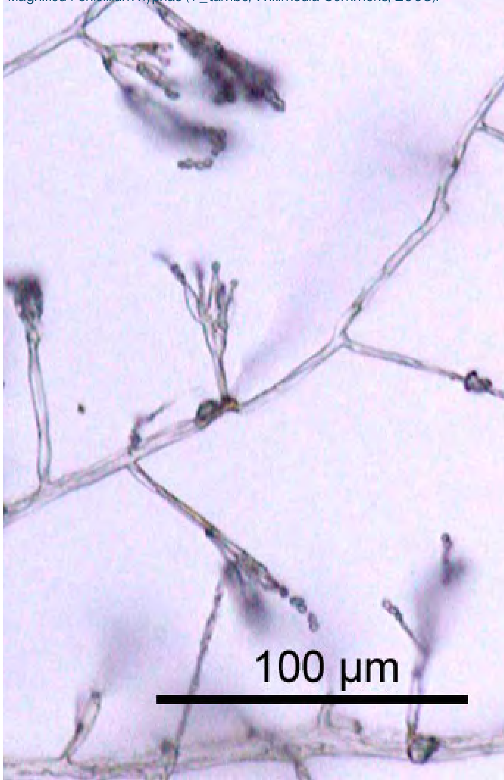
There are several types of wood-decaying fungi. In our context we are interested in the ones called «white-rot» fungi which belong to the basidiomycetes family. Wood-decaying fungi are decomposing and feeding on what basically composes wood: cellulose, hemicellulose and lignin. Given time, white-rot fungi will completely degrate lignin within the substrate they grow on, they also have particularly active enzymes which can degrade cellulose. White-rot fungi will prefer decidous trees wood to conifer's.



Structure of a multicellular fungus (Pearson Prentice Hall, 2004).

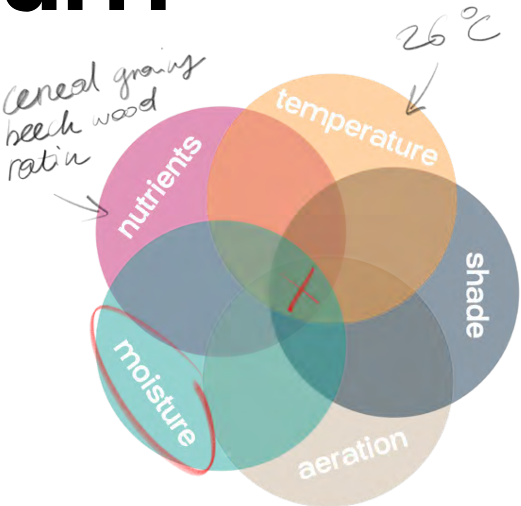
That is a mycelium too (Max.kit, Wikimedia Commons, 2016).

Magnified Penicillium hyphae (Y_tambe, Wikimedia Commons, 2005).



ganoderma resinaceum

Ganoderma resinaceum growing on a tree in Hong Kong (TuesdayHiker, Mushroom Observer, 2010).



Among the white-rot fungi species, *Ganoderma sessile* and *Ganoderma resinaceum* are some of the most «voracious» strains. These two are rather comparable in regards to their behaviours - as of the nutrients they feed on and the environmental conditions they foster with. *G. sessile* is more commonly found in North America, and *G. resinaceum* in Europe - we will work with the latter during this workshop.

When crafting the conditions for a mycelium to develop, we introduce good conditions for a lot of other microorganisms to develop too (such as other fungi and bacteria). To work around this contamination risk one can **sterilize the substrate** to work with, the **materials** in contact with the substrate, and the **work environment**. If the sterility is not ensured, then microorganisms will start to develop and ultimately compete with one another for the ressources in presence. This is one reason for us to chose to work with *G. resinaceum*, as it is a voracious strain it will perform better in presence of a contamination - and the development time of its mycelium is one of the fastest among white-rot fungi.

The scheme above is displaying the main parameters that one needs to ensure to foster fungal growth. Firstly, **myceliums develop in the shade**, away from UV exposure; for that reason, all of the experiments are kept away from direct light. Then we need to **ensure an access to air** for the fungi to «breath». This can be done when packing the substrate if it is not fully compressed, or by intruducing small perforations in the container holding the substrate. *Ganoderma* species tend to be performing better at a **temperature of about 26 degrees Celsius**. Then, the **substrate** that we have been over mentionning is actually the **source of nutrients for the fungi**. In our case we will be using a curation of nutrients: cereal grains, beech wood, and ratin fibres of various diameters.
Finally, the **moisture** is probably the most ephemerous and complex of the parameters we need to face; for the fungi needs access to the most water possible, without «drowning». The modelling activity will then focus on this aspect in workshop 2.

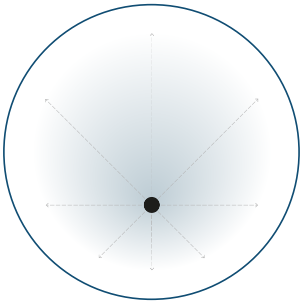
moisture model
/phenomenon

As we are interested in building a better understanding of the role that moisture dynamics play in the mycelium colonization of a substrate, we have reduced the variability of the other aforementioned parameters, or even fixed them (the shading is going to be constant as an example).
Now, we can hardly go look into the substrate to see how the moisture is arranged at a given time of the experiment...

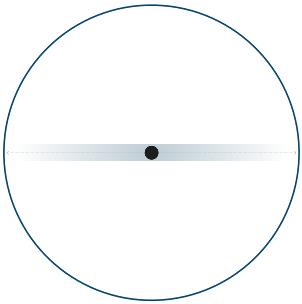
But we can identify the two main water phenomena at play as:

- the **evaporation** of the moisture that we initially introduced in the substrate,
- and the **capilarity** of the fibres.

In this model, we are not taking into consideration other phenomenon such as the influence of the mycelium itself over the distribution of moisture. The simulation workflow is described next page, where within the container's mesh, we are parsing random points all over the volume. these points serve as a discretization of the model; that is to say, the simulation will be only as accurate as the number of points we generate. Depending on your laptop's performances, we recommend a point quantity between 500 and 5000.

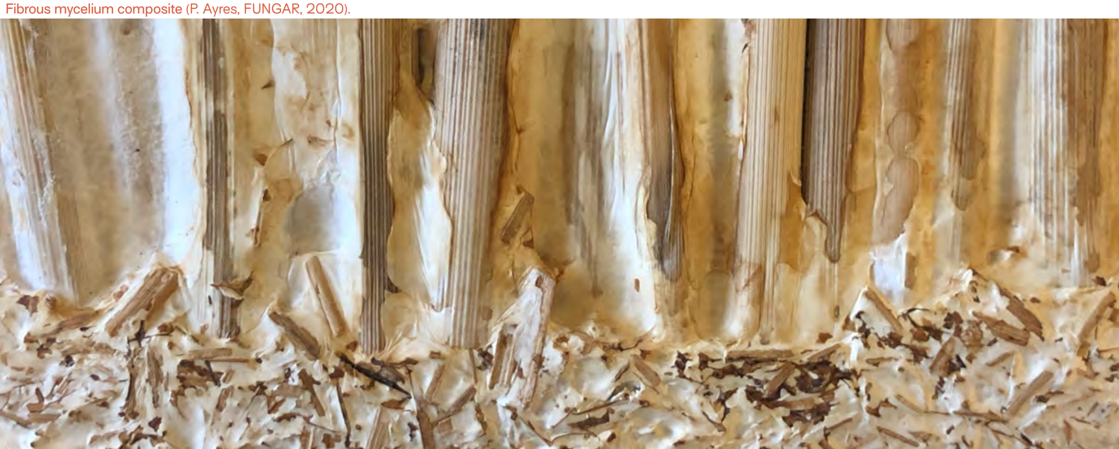


For the modelling purpose, we consider the **evaporation** to be sending vapor in all directions from its origin point, in an excentric fashion.

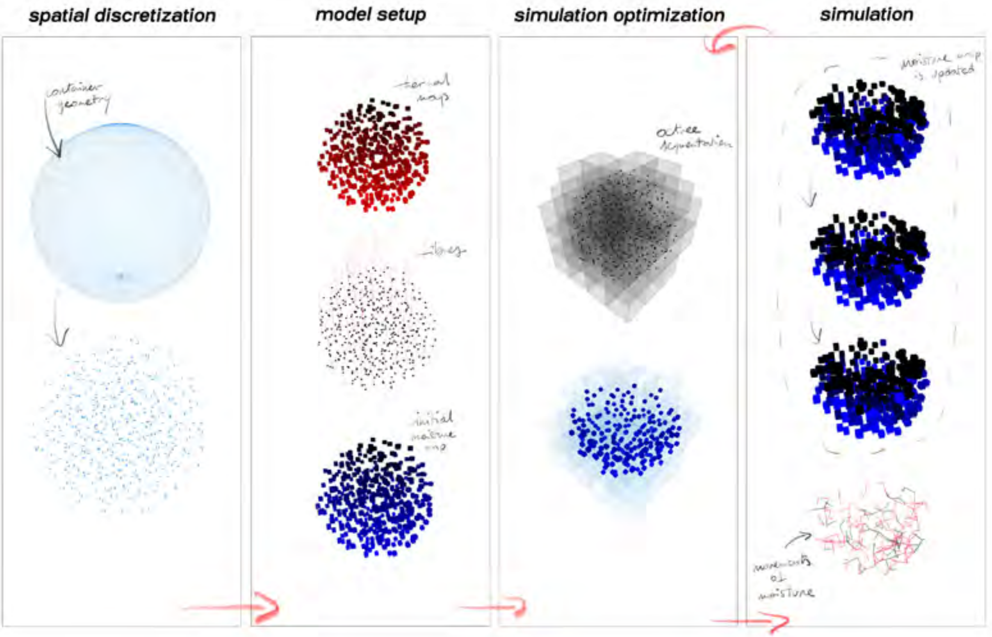


Capilarity is driving liquid water both ways along the axis of a fibre.

Fibrous mycelium composite (P. Ayres, FUNGAR, 2020).



moisture model /simulation workflow



moisture model /R&B

Once the model has been discretized, we need three more information to set it up: an **initial distribution of moisture**, a **distribution of the warmth** for the evaporation and capilarity intensities, and the **location of the fibers**. In this model, the total amount of moisture is considered constant.

The initial distribution of moisture needs to be formatted as an RGB Color value, on the Blue channel. The interval for this data type is [0; 255]. The warmth distribution is then formatted in a similar fashion, on the Red channel, again within an interval of [0; 255].

The model that we are working with comprises another layer of information; **for each value assigned in the model, a probability is assigned.** These probabilities represent an **indice of confidence** in the data being handled. As an example, in the picture below, we can know with precision the position of the long straw fibres on top, but we can only randomly guess how the small fibres of the bottom part of the experiment are organized.
Therefore, the longer fibres have a higher probability assigned to them.

Probabilities values are passed on with the Blue (moisture) and Red (warmth) data, following these relations (correspondance tables are on the next page):

- Moisture; $M_n = (B_n - 5 * \mu_{Mn}) / 5$
  with,
  B_n as the Blue value in [0; 255],
  M_n as the moisture level in [0; 50],
  μ_{Mn} as the confidence in the moisture level in [0.2; 1.0].
- Warmth; $W_n = (\mathrm{ceil}(R_n / 10.2) - 5 * \mu_{Wn}) / 5$
  with,
  R_n as the Red value in [0; 255],
  W_n as the warmth level in [0; 4],
  μ_{Wn} as the confidence in the warmth level in [0.2; 1.0].

For instance, if for a given point in the discretized space we want to set the maximum moisture level (50), and if we are fully confident that it is a valid level, then should set the Blue value as 255. If we are only 40% confident in this value, then we will set it to 252. The main difference in formatting between the Blue and Red datasets is that we are only taking into consideration five approximate levels of warmth. The correspondance between data and probabilities is working in a same way as for the Blue, with the addition of a remapping of the data with the division by 10.2, and an upper rounding of the division result (*ceil()* function).

Fibrous mycelium composite (A. Rigobello, FUNGAR, 2020).

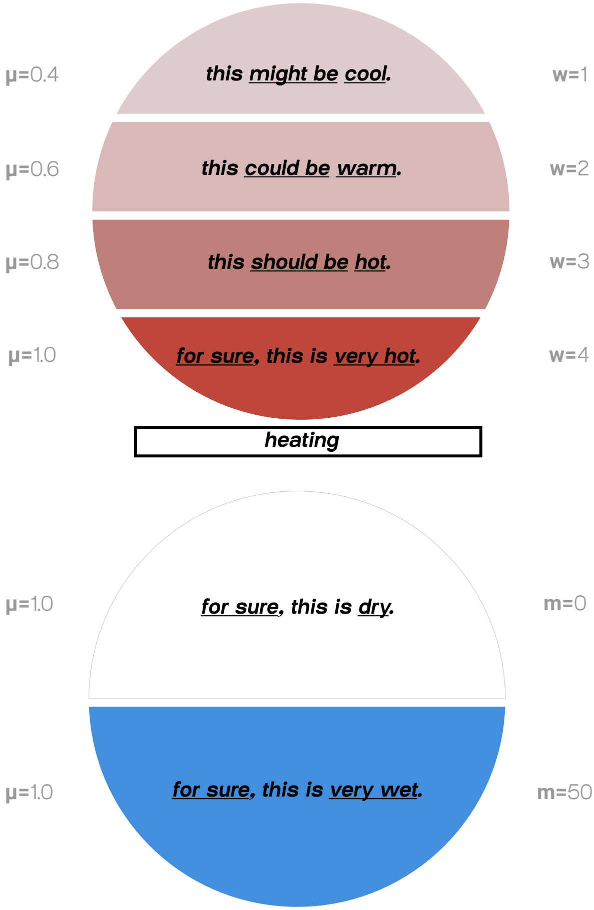
moisture model /R&B

Blue value (B_n)		Confidence in moisture level (μ_{Wn})				
		0,2	0,4	0,6	0,8	1
Moisture level (M_n)	0	1	2	3	4	5
	1	6	7	8	9	10
	2	11	12	13	14	15
	3	16	17	18	19	20
	4	21	22	23	24	25
	5	26	27	28	29	30
	6	31	32	33	34	35
	7	36	37	38	39	40
	8	41	42	43	44	45
	9	46	47	48	49	50
	10	51	52	53	54	55
	11	56	57	58	59	60
	12	61	62	63	64	65
	13	66	67	68	69	70
	14	71	72	73	74	75
	15	76	77	78	79	80
	16	81	82	83	84	85
	17	86	87	88	89	90
	18	91	92	93	94	95
	19	96	97	98	99	100
	20	101	102	103	104	105
	21	106	107	108	109	110
	22	111	112	113	114	115
	23	116	117	118	119	120
	24	121	122	123	124	125
	25	126	127	128	129	130
	26	131	132	133	134	135
	27	136	137	138	139	140
	28	141	142	143	144	145
	29	146	147	148	149	150
	30	151	152	153	154	155
	31	156	157	158	159	160
	32	161	162	163	164	165
	33	166	167	168	169	170
	34	171	172	173	174	175
	35	176	177	178	179	180
	36	181	182	183	184	185
	37	186	187	188	189	190
	38	191	192	193	194	195
	39	196	197	198	199	200
	40	201	202	203	204	205
	41	206	207	208	209	210
	42	211	212	213	214	215
	43	216	217	218	219	220
	44	221	222	223	224	225
	45	226	227	228	229	230
	46	231	232	233	234	235
	47	236	237	238	239	240
	48	241	242	243	244	245
	49	246	247	248	249	250
	50	251	252	253	254	255

Red value (R_n)			Confidence in warmth level (μ_{Wn})				
			0,2	0,4	0,6	0,8	1
Warth level (W_n)	0	Very Cold	10	20	31	41	51
	1	Cold	61	71	82	92	102
	2	Cool	112	122	133	143	153
	3	Hot	163	173	184	194	204
	4	Very Hot	214	224	235	245	255

moisture model
/R&B

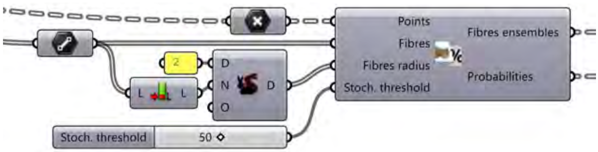
So to build the model correctly according to the previous rules, one can see the setup as such:



For instance, in the warmth map illustration above we can discretize the volume with our understanding of the warmth within the experiment. If we have a heating below the experiment, then we can be sure that the lower part is very hot, which corresponds to a **Warmth level of 4**, and a **confidence (Probability) of 1.0**. Therefore, the **spreadsheet on the previous page** gives us a corresponding **Red value of 255**, and we need to affect that value to the corresponding points. The principle is the same for the moisture map.
Please note that in the simulation **the total amount of moisture that is given as an input will be constant throughout the simulation.** It means that if every point has the minimum or the maximum moisture level, the simulation won't display any moisture dynamics. Similarly, if the warmth is low where the moisture is, very little moisture dynamics will be displayed.

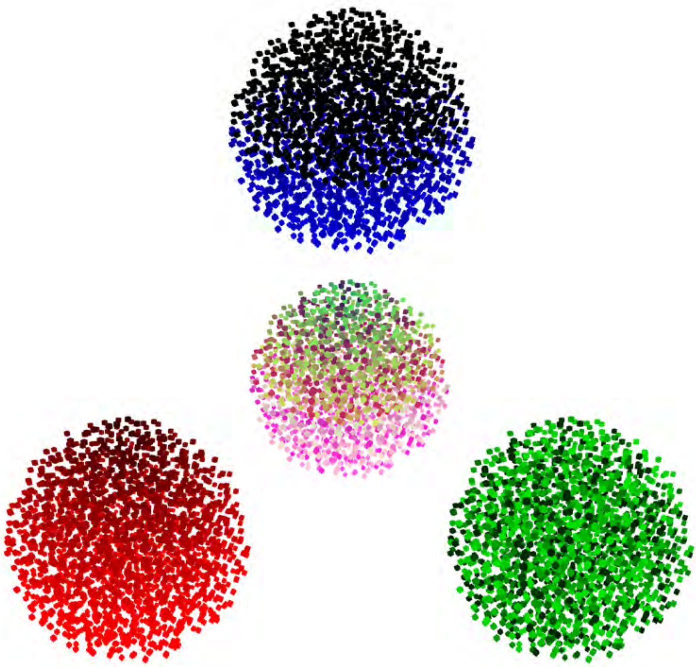
moisture model
/fibres

The third information that we need to provide the model with, is the **location of the fibres** in the model. To do so, the «Green generator» component of the «P. stellaviatori» plugin clusters points from the discretized model space by proximity to a previously drawn fibre.



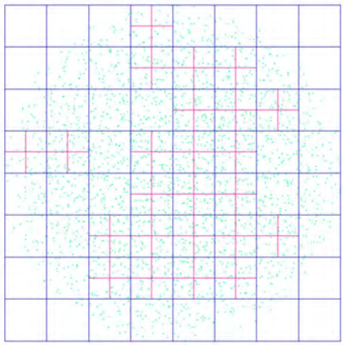
This component takes fibres as lines. The same amount of fibre radiuses as the fibre lines needs to be provided to the component. The «stoch. threshold» parameter sets a level of confidence in the fibre presence by comparing to the fibre line length. In the example above, each line that has a length of more than 50mm will be assigned a probability of 1.0. Conversely, the component assigns a probability of 40% (0.4) to fibres having a length in between (50/2)=25mm and 50mm, and 20% (0.2) under 25mm.

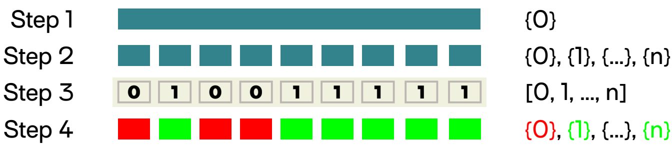
The illustration below displays a set of points with the three datasets rendered as RGB colors. Now that these datasets have been assigned, we will look at optimizing the workflow so to be able to increase the resolution of the model (i.e. the number of points).



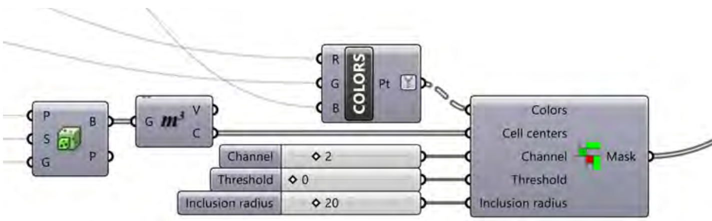
moisture model
/oc-tree

In order to optimize the simulation, we are using an oc-tree segmentation of the set of points. The oc-tree segmentation divides a volume in parallelepipedes, often cubes. Each subdivision results in the generation of 8 sub-partitions of a cube. While setting up the segmentation step, one needs to set the maximum number of points allowed in one subdivision. In the example aside that is in front view, the set of points in light green is segmented into parts of maximum 10 points. We can observe the presence of pink segments, which represent the smallest partitioning issued from the oc-tree process in this example. Then, after having generated the oc-tree cells in the definition, we are testing the inclusion of the points within each cell, and we are parsing them in branches of a datatree.



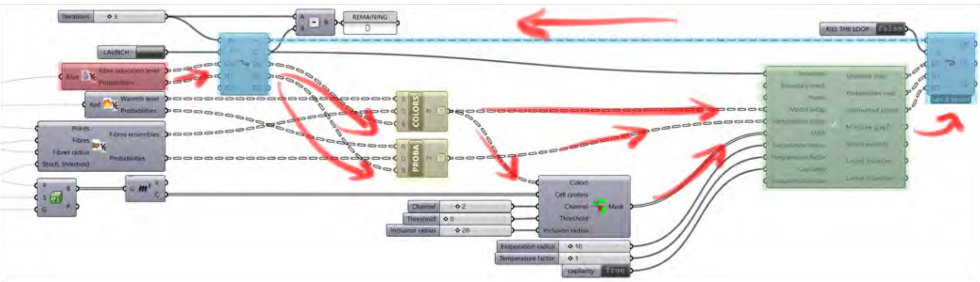


If we are taking a step back, at the initial stage (step 1) the points that we are parsing are all «flattened», at the same address {0} in the datatree. Then we test the inclusion of the points with the oc-tree segmentation cells; points are separated into groups, and are being affected a corresponding address {x} in the datatree (step 2). At the same time, Red and Blue colors that have been previously affected to points are restructured so that their datatree corresponds to the points' one. Then the Red, Blue and Green datasets are being aggregated along with the Green (fibres ensembles) and fed to the «ROI mask» component of the «P. stellaviatori» plugin. This is step 3, where the mask looks at the component's parameters and produces a boolean mask for the points groups that are relevant for the solver that comes afterwards. For instance, in the illustration below we are looking at the **Channel** 2 (Blue), and the component sets a «true» value to a given group is their average Blue value is above the **Threshold** value. As the solver goes on it might be sending moisture to points that did not previously have some. For this reason, we want to include points groups that are close to the ones that have moisture.
The component looks at the **Cell centers** and sets a «true» value to those within the **Inclusion radius**, measuring from cell centers that have moisture.



moisture model
/solver

Now that we have setup the inputs and covered the ROI mask component, this is all feeding into the **Solver** component. From the inputs, the solver (the green component on the picture) will iteratively look at updating the moisture map that is given depending on both the warmth map, and fibres ensembles (evaporation and capilarity). The probabilities affected to each of the inputs are also being aggregated by the solver.
The two blue components highlighted on the picture are from the Anemone plugin. They are allowing to build a loop with every component that is connected in between them. Here, the blue component on the left takes as D0 and D1 inputs the moisture map from the «Blue translator» component highlighted in red. The D0/D1 outputs of this left blue component, at the first iteration, are what is being fed with the Blue translator. D0, the moisture map, is being packed into the yellow highlighted component called «COLORS». D1, the moisture probabilities, is doing the same with «PROBA». Colors and Proba are then fed to the solver. Colors are fed to the mask too, as seen on the previous page. The solver sends the updated moisture and probabilities maps to D0/D1 on the blue component on the right, which then loops it to the component on the left - which updates the maps for the next solving iteration. This looping process is repeated N times (5 in this example), depending on the value of the slider in the upper left corner of the picture

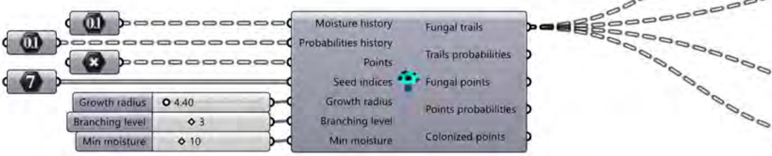


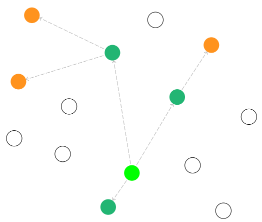
The solver also takes parameters such as the **Evaporation radius**, **Temperature factor**, and **Capilarity**. Capilarity is easy: this is a boolean to activate the simulation of fibres capilarity. The temperature factor simply boosts the simulation of both the evaporation and capilarity. The evaporation radius is the maximum distance at which a moisture-containing-point will send some evaporation at. It is not the same radius as the one of the ROI mask component; the ROI mask looks at distance between oc-tree cell centers, and the radius of the solver looks at the distance between points in the model.

Finally, after all the iterations have been completed, the D0 and D1 outputs from the blue component on the right are the list of moisture maps at each iteration (D0), and same for the probabilities (D1). It means that both output the complete history of the moisture / probabilities for the N iterations of the simulation.
As we can now see, the masks that we are using to optimize the simulation are changing throughout the simulation because they are embedded in the loop. So as to keep track of which points are considered at a given iteration, the D2 output on the right gives us a list of them for each iteration.

fungal trails

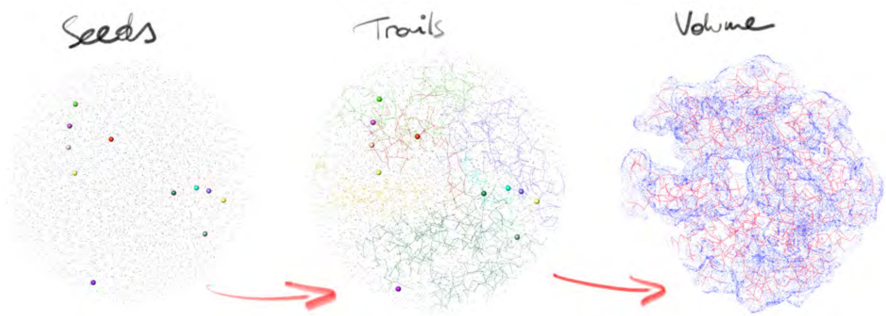
Once the moisture simulation completed, we basically have generated a list of moisture maps representing its evolution in time. Whith the results we can now simulate how mycelium will colonize it with the «DT trails» component of the «P. stellaviatori» plugin. As illustrated below, the component takes the **Moisture** and **Probabilities histories**, the list of **Points** that we have generated in the very beginning, and the indices of the inoculation points (**Seed indices**). These indices are pointing to the Points list.
In order to propagate from the seed points, the component looks into the neighboring points within the **Radius** if their moisture is enough (more than **Min Moisture**) for the mycelium network to extend. The Branching level parameter is basically a maximum of eligible neighboring points for growth. This means that a point can have up to 3 neighbors, or connections, in the example below.

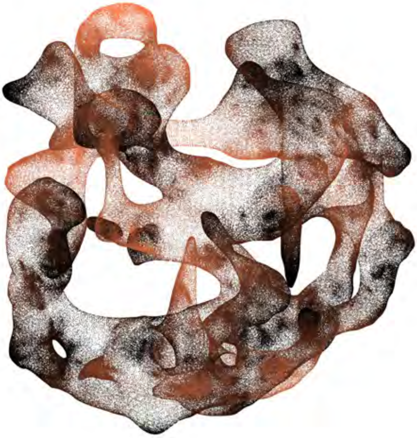


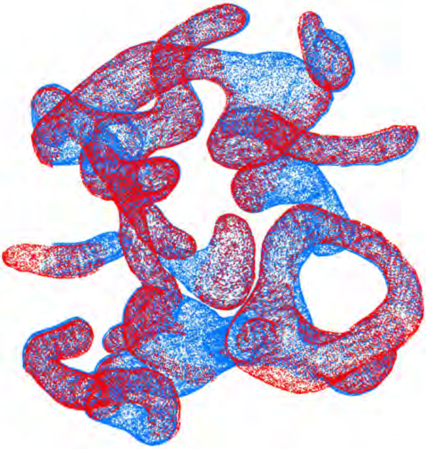
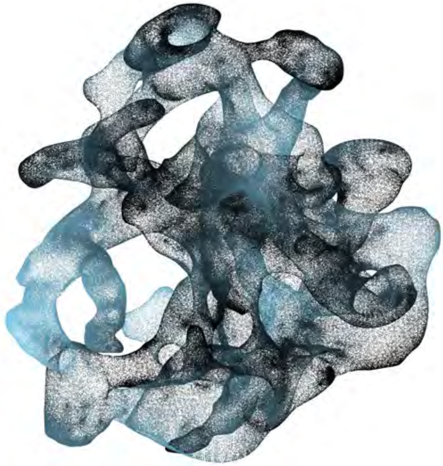
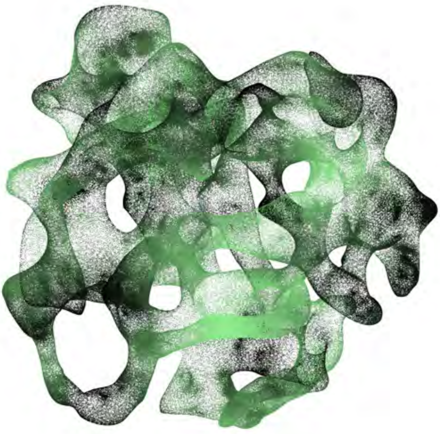


If the seed point is the light green one, it can find up to 3 neighbors (dark green), then at the next iteration, each of the dark green points will try to have up to 3 neighbors, but it might be that some cannot propagate further, or connect with less than 3 neighbors (Orange).

The graphs (trails, paths,...) that are being generated <u>do not</u> correspond to hyphae's scale, they are aiming at predicting mycelial growth at a larger scale. So from those trails, we can (and we will) generate volumes as meshes, and work around the various sets of information that we have been producing throughout the simulation with the goal to both inform the experimental setups, and explore those datasets to understand what we are really looking at.






Sterilization of substrates

Date: 09/05/2020
Version: 1.0
Author: Adrien Rigobello
Affiliation: CITA, KADK.

Abstract
This protocol describes the process to sterilize substrates for fungi cultivation, with a pressure cooker. This protocol applies to the sterilization of dry and wet substrates.

Materials

- Autoclavable bag with HEPA filters
- Curated substrate
- Tap water

Equipment

- Pressure cooker
- Electric cooking plate
- Electronic scale

Procedure

1. Weight the substrate to be sterilized and take note of the measure.

2. Weight the tap water to be sterilized and report it. The water mass should be a maximum of 90% of the substrate mass, and adjusted depending on its nature.

3. Fill the recipient (autoclavable bag), and stir to mix the water and substrate.

4. After having poured about 4 centimetres of water at the bottom of the pressure-cooker, place the recipient in the pressure-cooker, above the bottom of cooker (use a metallic basket or equivalent).

5. Close the pressure-cooker tightly, and set the pressure control to 60kPa.
   Set the electric plate on.

6. When the pressure cooker starts to blow, set a timer for 30 minutes of sterilization.

7. After the sterilization, stop the heating plate, release the pressure with the knob on the cooker, and carefully open it.

8. Carefully remove the containers from the pressure cooker and place them on a heat-compatible surface to let them cool down at ambient temperature. Dispose of the remaining water.

9. Once the substrates cooled down, use them in a sterile environment to avoid contamination.

Reference

Appendix

The F0 value is used to determine the exposure time of material for sterilization at a particular temperature, with a reference point be set at 121.11°C (250°F). The F0 value expresses the time in minutes for the specified temperature that gives the same thermal lethality as 121.11°C does in one minute.

$F0 = \Delta t * 10^{((T - 121.11)/Z)}$

With,
T = sterilization temperature,
Δt = 1min,
Z = 10°C.

With a pressure cooker at 117°C (60kPa):

$F0 = 1 * 10^{((117-121.11)/10)}$
$F0 = 10^{(-4.11/10)}$
$F0 = 10^{-0.411}$
$F0 = 0.388$

At 117°C, the lethality of the sterilisation is 0.388 times as effective as 1 minute at 121°C.
1/F0 = **2.58min** is the sterilization time required to be as effective at 117°C, than in 1min at 121.11°C.

Considering the fact that wet steam needs to reach each surfaces of the material to sterilize, the sterilization time is set as **30min**, which corresponds to a theoretical F0 of 11.63.

The Z coefficient (expressed in °C) characterizes the temperature variation modifying a micro-organism resistance of a factor 10 (1 log). The reference Z value for wet steam sterilization is the one of *Geobacillus stearothermophilus* which corresponds to Z ≈ 10°C.

Inoculating cellulosic substrates for fungi cultivation

Date: 09/05/2020
Version: 1.0
Author: Adrien Rigobello
Affiliation: CITA, KADK.

Abstract
This protocol describes the process to inoculate substrates for fungi cultivation.

Materials
- Sterile cellulosic substrates
- Spawn grown mycelial strain
- Optional: tap water
- Gloves

Equipment
- Bunsen burner
- 93° alcohol
- Electronic scale
- Stainless steel spatula

Procedure

1. Clean your workspace with the alcohol so to reduce the contamination risk by dust.

2. Clean the container in which you will inoculate with the alcohol.

3. If you are not working under a laminar flux, light up a Bunsen burner. The flame creates a sterile zone of a radius of 30cm, which will be your working area.

4. Weight the substrate to be inoculated and take note of the measurement.

5. Place the substrate in the clean container.

6. If the substrate is dry, add an equivalent of 90% of its mass in tap water, and stir with a clean spatula.

7. Weight an equivalent of 20% of the resulting mass in mycelium spawn.

8. With alcohol clean gloves, grind the mycelium spawn by hand in about 5-10mm big pieces. Alternatively, a clean blender can be used.

9. Add the mycelium to the container mix, and stir.

10. Close the container, and inoculate at the recommended temperature for the strain that you are using.

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D Journal articles

D.1 Compressive Behaviour of Anisotropic Mycelium-Based Composites

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Compressive behaviour of anisotropic mycelium-based composites

Adrien Rigobello✉ & Phil Ayres

Mycelium based composites (MBC) exhibit many properties that make them promising alternatives for less sustainable materials. However, there is no unified approach to their testing. We hypothesise that the two-phase particulate composite model and use of ASTM D1037 could provide a basis for systematisation. An experimental series of MBC were produced using four substrate particle sizes and subjected to compression testing. We report on their effect over Young's modulus and ultimate strength. We extend the study by investigating three anisotropic substrate designs through orientated fibre placement as a strategy for modifying compressive behaviour. We find that the two-phase particulate model is appropriate for describing the mechanical behaviour of MBC and that mechanical behaviour can be modified through anisotropic designs using orientated fibres. We also confirm that fibre orientation and particle size are significant parameters in determining ultimate strength.

Mycelium-based composites (MBC) are being investigated in design and materials engineering by leveraging the saprotrophic lifestyle of ligninolytic fungi, taking inspiration in the XIXth to early XXth century method of fungal strain transfer by lignocellulosic solid-state cultivation[1]. Because MBC cultivation protocols can be based on virtually any substrate containing organic polymers such as lignin, hemicellulose and cellulose, and as they instrumentalise a range of widely available basidiomycota, this class of composite shows potential in obtaining viable products for a variety of uses. Furthermore, MBC conform to circular economy production principles, are expected to be biodegradable, and are assumed to have a low environmental impact in regards to Life-Cycle Assessment (LCA). Lignocellulosic substrates cover a variety of geometries and chemical profiles, from industrial grade dusts and particles to supplies of irregular shavings, from grain husks to straws; this variety of supplies has led to the emergence of a rich craft in MBC production. However, this poses a challenge in systematically understanding the behaviour of this new class of materials. We argue that rationalising and systematising approaches to analysing their complexity is necessary to actualise their potential and facilitate market readiness.

No analytical model has been previously proposed for MBC. We hypothesise that they qualify as two-phase particulate composites with the fungal mycelium acting as the matrix, and the substrate, with a high particle content ratio and randomly orientated, acting as the dispersed phase. Because the mechanical response of the fungal mycelium that binds particles together acts as a foam[2], the composite stiffness is primarily driven by the substrate composition with angular particles. Previous studies of the failure mode of two-phase particulate composites have extensively investigated particle dewetting and their interfacial interactions for high particle content ratios[3]. Fourier-Transform Infrared (FTIR) spectrometry was used to qualify the materials used as principal substrate and fibre addition, the mycelium of *G. lucidum*, and *G. lucidum* colonised beech wood. Th s study then focuses on the influence of particle size on the compressive behaviour of MBC. The study is then extended to examine the influence of orientated fibres for modifying mechanical behaviour through anisotropic design. Three granulations of beech wood from 0.5 to 12.0 mm are used.

Materials and methods

Standard reference for specimen design. A variety of experimental designs are being used in the fi ld of MBC research and engineering, as there is a current lack of unifi d approach to the material description. Few studies consider evaluation standards for MBC; among them, ASTM D3501 for wood-based structural panels in compression has been referenced[4], a standard designed for plywood, wafer-board, orientated strand board, and composites of veneer and of wood-based layers, with use of 2:1 (D:h) cylindrical specimens in the study, instead of rectangular cross-section as the standard advises; ASTM D695 for rigid plastics was also referenced[5], with a recommended 1:2 (D:h) ratio for cylindrical samples, but used with a diameter of 100mm and thickness of 23mm in the study; ASTM C67 destined to brick and structural clay tile was referenced in a comparative study

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against clay bricks[6], but without following the standard recommendations; and ASTM D2166-13 for cohesive soil was referenced[7], but deviating from the standard in the study. In requiring the largest particle to be smaller than one tenth of the specimen diameter, the latter ASTM exemplifies the instrumental role these standards can play in systematically investigating materials based on previous studies. The variety of specimen geometries found in the state of the art and lack of consistent recourse to the standards, challenges the portability of, and comparability between, experimental results.

Hypothesising that the best fit model for MBC is as two-phase particulate composite with a high particle content ratio, with particles randomly orientated, and, for this study, with sizes in the 0.5–12.0 mm range, we identify ASTM D1037 for assessment of wood-base fibre and particle panel materials mechanical properties as the most appropriate material standard for specimen and experimental plan design. We report compression parallel to surface evaluation, for which the short-column method has been chosen as the specimens have of a nominal thickness above 25 mm. They are parallelepipeds of 1:1:4 ratio, the nominal dimensions are $36 \times 36 \times 144$ mm and the dimensions of the dried specimens are $34 \times 34 \times 140$ mm. Our experimental plan investigates the effect of granulate sizes and reinforcement strategies over the compressive Young's modulus and ultimate compressive strength.

Principal substrates. The principal substrates of the specimens originated from European beech wood (*Fagus sylvatica*). To investigate the effect of particle sizes over the compressive behaviour, we used three granulations (small, medium, large): 0.5–1.0 mm (Räuchergold type HB 500/1000, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany), 0.75–3.0 mm (Räuchergold type HB 750/2000, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany), and 4.0–12.0 mm (Räuchergold type KL 2/16, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany). A fourth particle type was added to the experimental plan, as a 1:1:1 volume ratio mix of the three granulations.

Fibre compositions. Longitudinal fibres were introduced in a specimen series by using common reed fibres (*Phragmites australis*; Tækkemand Chresten Finn Guld, Køge, Denmark). Eight to ten fibres of 1 mm ± 0.5 diameter were chosen so as to balance their dimensional variability and positioned in two layers separated by 10 mm of principal substrate. Fibres perpendicular to compressive stress were studied with use of 6 mm diameter by 32 mm length rattan fibres (*Calamus manan*; B.V. INAPO, Bloemendaal, Netherland). They were positioned regularly within the principal substrate as two layers of fibres, centred in the specimen thickness and separated by a 10 mm layer of principal substrate. It is common in MBC design practices to have mycelium grown externally on the outer boundaries of the specimens[4]. In the context of this study, no external mycelium was grown so as to observe the effect of granulate sizes and reinforcement strategies without introducing a specimen geometry bias. We identify this bias as critical for the reproducibility of experiments as the characteristics of the external mycelium mat is never found to be reported in the state of the art. In this study, a jacketing strategy has been integrated to study the effect of boundary reinforcement with a reproducible method. Across granulate sizes, a hemp-based hessian jacket (*Cannabis sativa* subsp. *sativa*; NEMO Hemp jam web 370 g/m^2, Naturellement Chanvre, Echandelys, France) was introduced on the specimen length. The study complies with relevant institutional, national, and international guidelines and legislation regarding the use of plant materials.

Fungal species. *Trametes* spp., *Ganoderma* spp., and *Pleurotus* spp. are among the most frequently cited families in MBC design[8]; *Schizophyllum commune* is a less investigated species but fi ds a growing interest[9], and *Irpex lacteus* has been used previously[7]. 565 carbohydrate-active enzyme families (CAZymes) have previously been assigned to the *Ganoderma lucidum* species[10,11], representing the widest array from hydrolytic enzymes (hydrolysis of hemicellulose, pectin), to oxidoreductases (laccases, ligninolytic peroxidases and peroxide-generating oxidases), to cellobiose dehydrogenase. Because this species is considered a very versatile ligninolytic fungus, in that it can exploit various strategies for the breakdown of lignin and can ultimately degrade all components of lignocellulosic compounds, it was selected for implementing the experimental plan. A millet-grown spawn of ligninolytic species *G. lucidum* (reference M9726) was acquired from Mycelia BVBA (Nevele, Belgium). The spawn was stored at a constant 4 °C and 65% relative humidity (RH).

Specimen preparation. The moisture content (MC) within cell walls as bound water, and outside cell walls in wood void structure as capillary water or vapour, is critical for understanding and predicting fungal activity[12]. In MBC production, lignocellulosic substrates composed of particle or fibres have a MC that is homogeneously prepared at 55–70%[4,13], and the use of mineralized to sterile demineralized water has been documented as a moisturising mean[4,13,14].

For this study, the principal substrates, fibres and hessian were prepared at 40% MC with mineralized water, and sterilised at 121 °C for 15 min. The principal substrates were then mixed with 16 wt% *G. lucidum* spawn and incubated in PP filtered bags (PPD50/REH4+1/V22-49, SacO2, Deinze, Belgium) for 7 days at 25 °C in the dark. Once colonised, the principal substrates were massaged to break them down and formed with the fibres and hessian into aerated PETG moulds. The formed specimens were incubated for 21 days at 25 °C in the dark, then oven dried for 48 h at 60 °C. The dried specimens were stored at 4 °C and 65% RH prior to testing.

Compressive behaviour characterisation. The use of seismic waves to characterise the mechanical behaviour of MBC has been reported in the literature as an alternative to conventional uniaxial load testing[7]. Th s method has become common in geological and civil engineering, and offers the benefit of being non-destructive. However, the anisotropic nature of the composite matrix (the mycelium), together with is its high elasticity, causes waves to attenuate irregularly. Furthermore, MBC have such a high porosity and a high variation

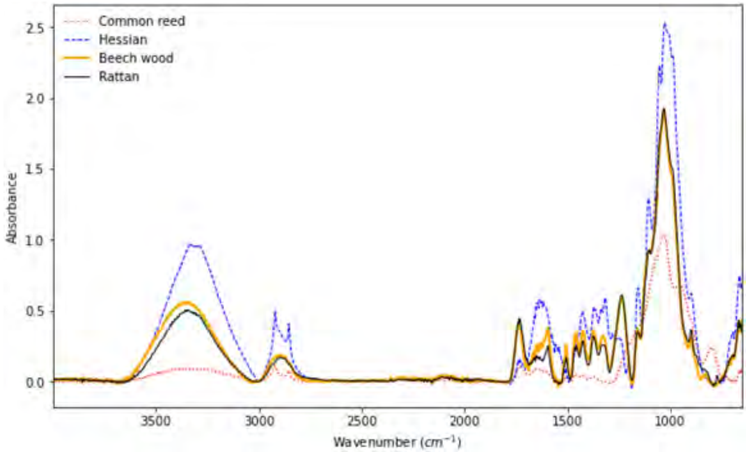

Figure 1. FTIR spectra of hemp-based hessian, beech wood, rattan, and common reed fibres.

in particle sizes and distribution that gaining accurate measurements would be challenging. Th s is evidenced in the literature by a larger standard deviation in results using this process applied to homogeneous MBC[7]. In the context of this study, load testing was performed on a Mecmesin MultiTest-dV testing bench equipped with a 2500 N load sensor, with a loading speed of 1.0 mm/min. Young's modulus and ultimate compressive strength were calculated following ASTM D1037.

Chemical analysis. Fourier-Transform Infrared (FTIR) spectrometry has been used previously for analysing the lignocellulosic profiles of substrates and their relation to fungal degradation patterns[4,15–17], with the benefit of requiring a limited specimen preparation, and spectra shape and frequencies being directly related to microscopical physical quantities and hence prepared for interpretation[18]. FTIR spectrometry was conducted in this study on a single refl ction diamond Attenuated Total Refl ctance (ATR) Agilent 4500a FTIR (Santa Clara, USA). The acquisition resolution was 4 cm^{-1} with 16 scans per specimen, for a band between 4000 and 650 cm^{-1}. We corrected the baseline of FTIR spectra following the adaptive iteratively reweighted Penalised Least Squares (airPLS) method[19], and spectra normalization was done with amide I/II band envelopes[20]. Four samples were isolated from *G. lucidum* colonised beech wood specimens, their spectra were averaged for analysis. The other specimens were tested with one replicate.

Chemical analysis

To serve as controls, we used FTIR spectrometry to characterise the four materials used in the composite design (Fig. 1). The materials were hemp-based hessian, beech wood, rattan, and common reed. Beech wood and rattan spectra display a chemical profile that is very similar, with the exception of peaks at 1123 cm^{-1} and 1160 cm^{-1}, and the 1300–1500 cm^{-1} region. Th s indicates a slightly higher content of cellulose, hemicellulose and lignin in our tested beech wood specimen (C–O stretching, C–O–C asymmetrical stretching, C–H deformation, COOH groups symmetrical stretching, symmetric C–H bending, CH_2 deformation stretching, CH_3 asymmetrical angular vibration, vibrational mode of amide C–O stretching)[21–23]. Common reed displays a minimal amount of lignin and hemicellulose compared to our other samples, while the peak at 890 cm^{-1} is associated with C–O–C stretching at the β–(1→4)–glycosidic linkages of amorphous cellulose[24]. The hessian displays distinctive peaks at 707 cm^{-1}, 890 cm^{-1}, 1060 cm^{-1}, 1316 cm^{-1}, and 1430 cm^{-1} in the fi gerprint region, and 1640 cm^{-1}, and 2921 cm^{-1}. The 750–680 cm^{-1} and 1680 – 1630 cm^{-1} regions (C=O streching) are associated with primary and secondary amides in hemp (amide V: C–N and N–H vibrations)[25]. Primary amides in hemp are amino acids, fatty acids, and steroids, which contribute to the 3500–3000 cm^{-1} region. The 1310–1230 cm^{-1} region (C–N stretching) is associated to secondary amides, such as cannabinoids, flavonoids, stilbenoids, terpenoids, alkaloids, and lignans[26]. The peak at 2921 cm^{-1} is associated with alkyl C–H groups[27].

Four samples were isolated from *G. lucidum* colonised beech wood specimens after they were used for load testing. Their spectra were averaged and are presented on Fig. 2 along with the beech wood spectrum, and a sample of *G. lucidum* mycelium. Peaks at 886 cm^{-1}, 1075 cm^{-1} and 1160 cm^{-1} are characteristic of (1→3)– and (1→6)–β–glucans that are present in the fungal cell wall (identified as [2], and [4] on Fig. 2). The peaks [1] and [3] at 780 cm^{-1} and 1043 cm^{-1} are also associated with β–glucans[28]. Chitin is identified at peak [5] 1313 cm^{-1} (amide III: C–N stretching), which also affects the 1640 cm^{-1} region [6] alongside the presence of peptides and

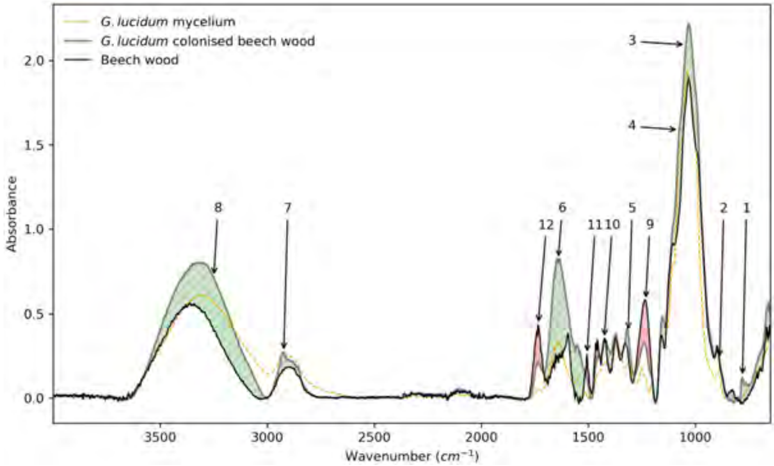

Figure 2. FTIR spectra of *G. lucidum* colonised beech wood, *G. lucidum* mycelium, and beech wood. Green areas represent increased values in mycelium-colonised specimens (peaks 1 to 8), red areas are decreased values in mycelium-colonised specimens (peaks 9 to 12).

secondary metabolites (aromatic rings and conjugated alkenes). The peak [7] at 2922 cm^{-1} is representative of chitin and ergosterol (C–H stretching)[29]. The 3600–3000 cm^{-1} region (peak [8]) is considered to be influenced by residual water and entrapped CO_2 (O–H and N–H stretching). Finally, peaks [9] to [12] represent decreases at 1231 cm^{-1}, 1425 cm^{-1}, 1506 cm^{-1}, and 1733 cm^{-1}. They are associated with lignin and xylan breakdown (syringyl ring breathing and C–O stretching, C=C stretching vibration in aromatic ring), and cellulose (peak [11]) and hemicellulose (peak [11] and [12]) breakdown is observed (CH_2 scissor vibration, C=O stretching)[30]. To evaluate the lignocellulosic changes undertaken during *G. lucidum* activity quantitatively, the band ratio indices at 1231 cm^{-1}, 1425 cm^{-1}, and 1506 cm^{-1} were calculated from the 2921 cm^{-1} band[31] for beech wood and *G. lucidum* colonised beech wood as:

$$\frac{I_n}{I_{2921}}, \tag{1}$$

Where I_n is the specific band intensity and I_{2921} the band intensity at 2921 cm^{-1}. The band ratio at 1231 cm^{-1} went from 1.58 in beech wood to 0.61 in *G. lucidum* colonised beech wood; the band ratio at 1425 cm^{-1} went from 0.96 in beech wood to 0.63 in *G. lucidum* colonised beech wood; the band ratio at 1506 cm^{-1} went from 0.71 in beech wood to 0.24 in *G. lucidum* colonised beech wood; the band ratio at 1733 cm^{-1} went from 1.19 in beech wood to 0.41 in *G. lucidum* colonised beech wood. We can therefore observe that *G. lucidum* had a preference in breaking down lignin and xylan at 1231 cm^{-1} compared to cellulose and hemicellulose at 1425 cm^{-1} (2.94:1), which is confi med by the ratios at 1506 cm^{-1} for lignin (1.42:1), and 1733 cm^{-1} for hemicellulose (2.36:1). The CH_2 scissor vibration corresponding to the peak at 1425 cm^{-1} refl cting both cellulose and hemicellulose, the present decrease might be primarily related to hemicellulose breakdown. Th s preference of *G. lucidum* for lignin and hemicellulose is consistent with fi dings reported in the literature[10].

Compressive behaviour

We investigated the effect of particle sizes on the mechanical behaviour in compression of MBC using four levels of granulation: small (BS family), medium (BM family), large particles (BL family), and a 1:1:1 volume ratio mix of the three previous granulations (BSML family). A second parameter was introduced to investigate the anisotrope modification of MBC. Three typologies of fibre composition were implemented in the experimental plan: hessian jacketing coaxial to the load case (H), unidirectional rattan fibres perpendicular to the load case (R), and unidirectional common reed fibres coaxial to the load case (V). Isotropic controls were added for each level of granulation (BS, BM, BL, BSML specimen types in the figu es). Fig.3 illustrates the three typologies alongside the control. Experimental parameters per specimen type and resulting mean density, mean Young's modulus and mean ultimate strength are presented in Table 1. Box plots of the results for Young's modulus and ultimate strength are presented in Fig. 4, and box plots for densities are reported in Fig. 5.

Specimen type	Granulate size (mm)	Fibre composition	Mean density (s.d.)	Mean Young's modulus (s.d.)	Mean ultimate strength (s.d.)
BS	0.5–1.0	Control	209.67 kg/m^3 (6.47)	1.79 MPa (0.41)	171.86 kPa (36.54)
BS_H	0.5–1.0	Hessian jacketing	230.48 kg/m^3 (9.88)	1.58 MPa (0.42)	175.79 kPa (34.38)
BS_R	0.5–1.0	Rattan perpendicular to load	196.59 kg/m^3 (19.01)	0.66 MPa (0.42)	89.06 kPa (58.49)
BS_V	0.5–1.0	Common reed coaxial to load	194.12 kg/m^3 (5.09)	3.88 MPa (2.51)	146.85 kPa (39.26)
BM	0.75–3.0	Control	233.87 kg/m^3 (9.04)	3.32 MPa (0.80)	306.38 kPa (57.64)
BM_H	0.75–3.0	Hessian jacketing	248.70 kg/m^3 (12.20)	2.99 MPa (0.54)	298.85 kPa (35.47)
BM_R	0.75–3.0	Rattan perpendicular to load	226.77 kg/m^3 (6.19)	4.02 MPa (4.45)	232.30 kPa (62.85)
BM_V	0.75–3.0	Common reed coaxial to load	198.14 kg/m^3 (2.89)	9.21 MPa (6.42)	270.93 kPa (79.76)
BL	4.0–12.0	Control	217.60 kg/m^3 (10.58)	2.96 MPa (1.04)	245.60 kPa (30.31)
BL_H	4.0–12.0	Hessian jacketing	264.05 kg/m^3 (11.97)	3.01 MPa (0.46)	223.93 kPa (25.88)
BL_R	4.0–12.0	Rattan perpendicular to load	240.98 kg/m^3 (3.91)	2.24 MPa (0.58)	180.88 kPa (64.75)
BL_V	4.0–12.0	Common reed coaxial to load	209.47 kg/m^3 (7.90)	8.50 MPa (4.56)	290.86 kPa (100.83)
BSML	0.5–12.0	Control	220.59 kg/m^3 (8.12)	2.17 MPa (0.36)	237.09 kPa (31.73)
BSML_H	0.5–12.0	Hessian jacketing	246.85 kg/m^3 (11.29)	2.20 MPa (1.04)	194.48 kPa (48.47)
BSML_R	0.5–12.0	Rattan perpendicular to load	224.09 kg/m^3 (3.55)	1.87 MPa (0.30)	171.44 kPa (26.13)
BSML_V	0.5–12.0	Common reed coaxial to load	203.08 kg/m^3 (6.24)	7.89 MPa (2.41)	338.75 kPa (65.39)

Table 1. Summary of specimen types parameters, resulting dried densities, and compressive properties.

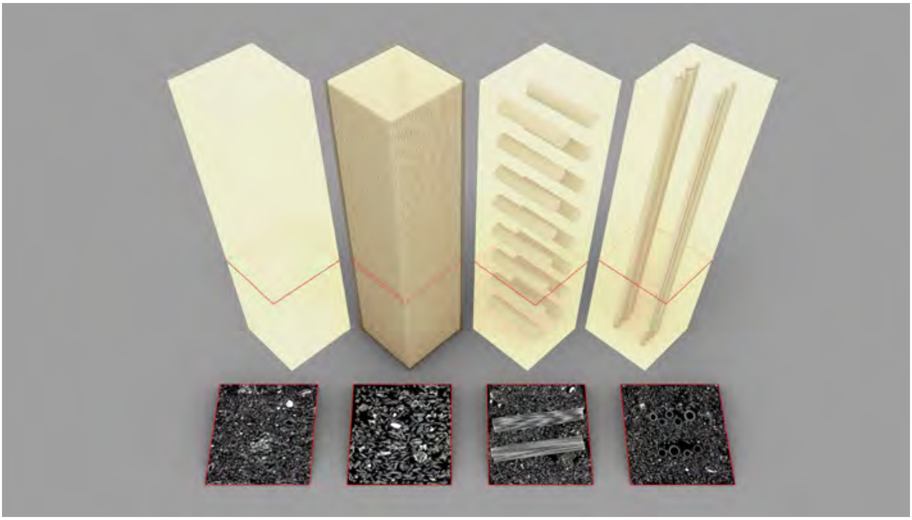

Figure 3. Fibre placement strategies and their sectional CT scan (left o right): control (BS), jacketing coaxial to load (BM_H), fibres perpendicular to load (BS_R), fibres coaxial to load (BS_V).

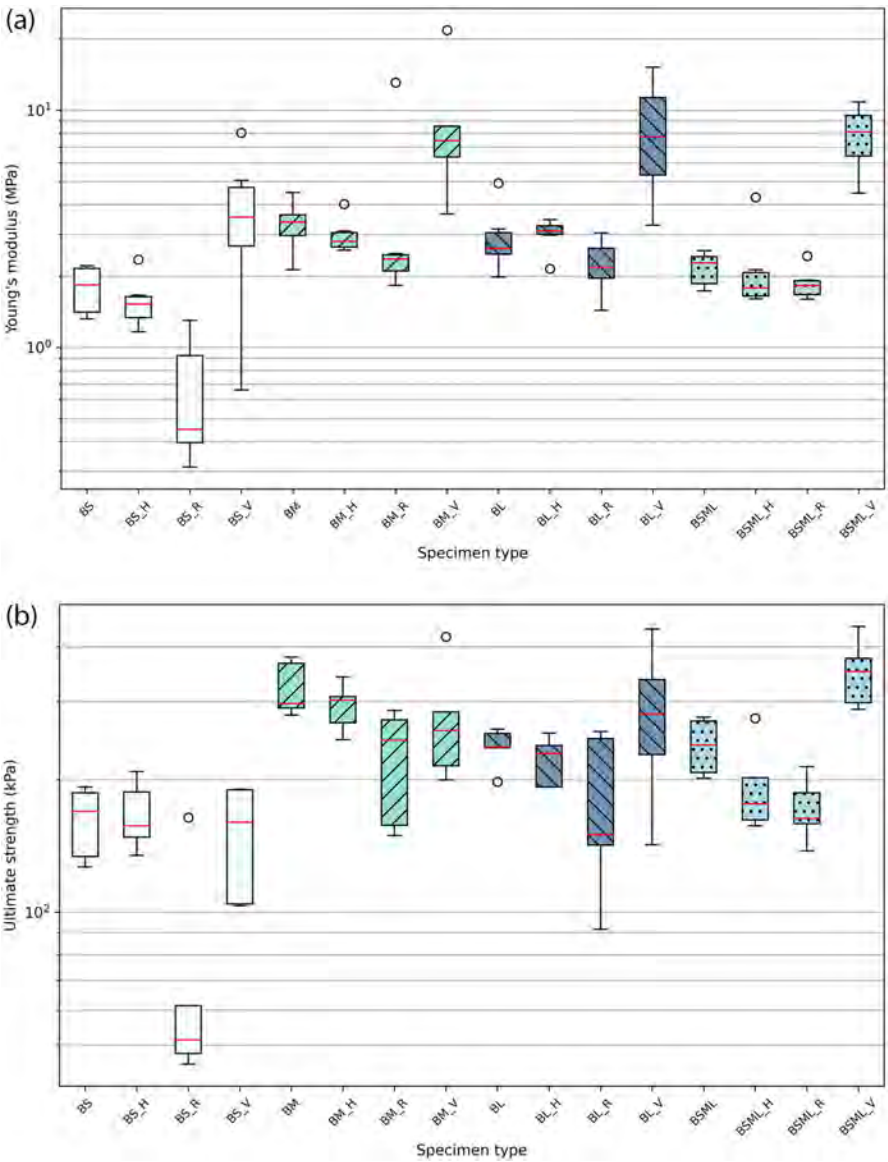

Figure 4. Box plots for Young's modulus results (**a**) and ultimate strength results (**b**).

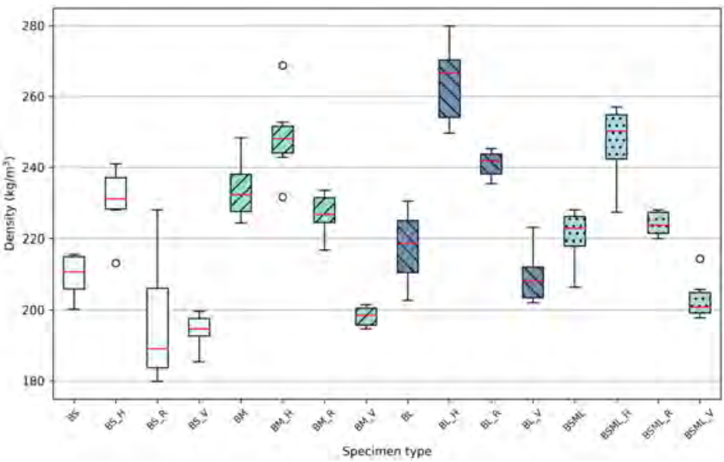

Figure 5. Box plots for dried specimen densities.

Jacketing coaxial to load. The introduction of the hessian jacket offers a contrasting illustration of the effect of the mycelial mat usually grown on the external boundary of MBC. We observe that the dispersion of Young's modulus results across all specimen families is reduced compared to their controls with the exception of the BL family. The jacketing also affects the dispersion of results in ultimate strength in the case of the BS, BM, and BSML families, with a reduction of the deviation between the fi st and third quartiles. The containment of stress applied to the specimens within tight boundaries forces the arrangement of the particles within, restricting the ability for particles to arrange freely. Jacketed specimens have an average reduction of 0.12 MPa to the controls as per Young's modulus (s.d. 0.19), and an average decrease of 16.97 kPa to the controls as per ultimate strength (s.d. 20.05). The jacket has two important advantages: it offers a durable alternative to low-ductility mycelial mats usually grown on the external boundary of MBC, and we hypothesise that it can substantially contribute to an increase in fracture resistance performance in shearing and bending load cases.

Fibres perpendicular to load. Specimens supplemented with rattan fibres display a lower performance across particle sizes considering their median in Young's modulus and ultimate strength. The mean ultimate strength follows the performance of the mean of the controls (Fig. 6) with an average reduction of 71.81 kPa (s.d. 8.45). Th s suggests that, should the production conditions of such MBC improve to reduce the dispersion of results and increase the material behaviour predictability, introducing strategically parsed weakness points in composites could fi d a use with calibrated materials by tuning their failure mode.

Fibres coaxial to load. Common reed fibre reinforced specimens resulted in the largest standard deviations in Young's moduli (reported in Table 1), especially in the BM and BL families. Th s is due to the fibres having partially misaligned to the load case axis during specimen production. Nevertheless, results suggest that MBC can be successfully stiffened with regards to their use case. The effect of this stiffening on the ultimate strength is less obvious as we note that the smaller particles (BS and BM families) tend to perform better without reinforcement coaxial to load. Th s is a result of the inherent large displacement of the fibres within the specimens under stress due to their stiffness, thus initiating an early critical failure. The mean Young's moduli (Table 1) display a clear improvement compared to the controls: we observe an average increase of a factor 2.86 (s.d. 0.6) between the mean of the controls and the mean of the fibre coaxial to load specimens. As per mean ultimate strengths, they improved in the BL and BSML families when compared to controls (respectively by a factor 1.18 and 1.43), but decreased in the smaller particles families BS and BM (respectively by a factor 0.86 and 0.88).

Principal substrate particles. The use of smaller particles in MBC increases the surface area to volume ratio of what serves as a nutrient for the fungus, hence facilitating its access to it. Fungi also need air access to develop a mycelium, and space between particles, if one desires to have it synthesise a biomass that has a considerable effect over its mechanical properties. The small granulation essentially qualifies as a dust with particles size in the 0.5–1.0 mm interval, leaving minimal amounts of air between particles within the constrained boundaries of the specimen mould. The best performing BM family (as per ultimate strength) is composed of 0.75–3.0 mm particles, thus embedding particles of a comparable size to the BS dust, while containing particles

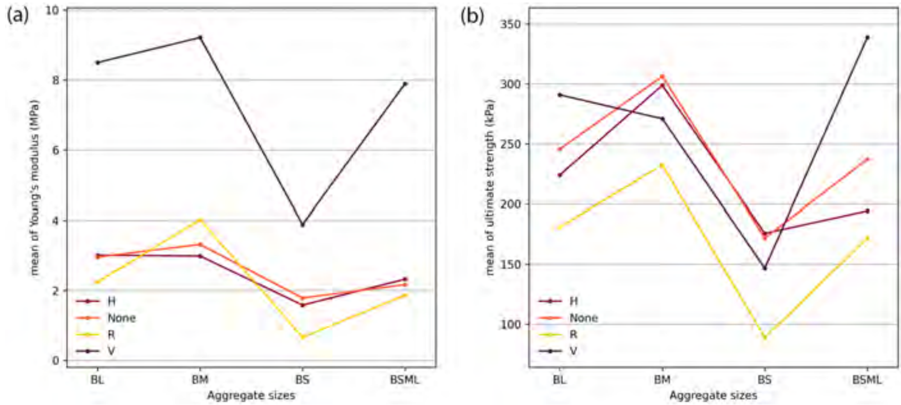

Figure 6. Parameters interaction graph for Young's modulus (**a**) and ultimate strength (**b**).

that are up to six times as long. Th s understanding is nonetheless challenged by looking at the BS, BM, BL and BSML group densities (Table 1 and Fig. 5), where the BS group has the lowest resulting density.

Studying a material model composed of cylindrical particles with a length in 2–10 mm and diameter of 0.5–2 mm, a study concludes that the matrix phase of MBC is ruling the composite modulus[2]. Th s study avoids considering more particle shape parameters that have been shown to have a signifi ant effect over the system behaviour; flakiness/flatness (thickness to width ratio), elongation (length to width ratio), sphericity (deviation from a sphere geometry), and roundness/angularity (angular sharpness) have been previously investigated in particle studies of granular materials[32]. It was reported that a 3:1 ratio of flaky particles content would be an approximate optimal for shear strength (depending on the system of study). This is related to cohesion being increased under stress due to particle interlocking. A higher particle angularity was reported to induce a decrease in elastic modulus, and an increase in ultimate strength. Shear strength was reported to increase with particle angularity. Increasing particle flakiness and angularity increases cohesion and abrasion. Th s leads to damage accumulation under repeated loads, resulting in strain accumulation[32]. This suggests that modifying composite behaviour does not only depend on mycelial expression, but should investigate substrate contribution too systematically.

While the BM group mean density is 11.54% higher and the most different to the BS group mean density, the BM mean elastic modulus is 85.48% higher than that of the BS group and 78.27% higher in ultimate strength. In this experiment, there is a correlation between an increased density and increased stiffness and strength. As the principal substrate used in these four groups is of the same nature and source, we can note that the particle volume fractions are directly correlated to the densities. It is worth noting too that the different levels of granulation result in different aggregate mechanical properties; on Fig. 7 we can notice the effect of comminution, smaller granulation (a) result in a higher content of short fibres with a lower bending stiffness, medium sized particles (b) display a content of not only fibre-type particles but also less elongated, more angular and bulky ones, contributing to increase their bending stiffness and interlocking potential under stress, and finally the larger granulates (c) display an increased flakiness and angularity to the medium ones. While the latter would be expected to result in higher stiffness and strength to the other groups because of their aggregate geometrical characteristics, the manufacturing of the specimens did not focus on particulate arrangements for this series and therefore these were randomly orientated and thus not optimised for interlocking.

Pure mycelium material response under tensile and compressive stress has been investigated and modelled[2], and has been classified as an open-cell foam-like material. Pure mycelium of an undisclosed species in this study was reported to exhibit a Young's modulus of 0.6–2 MPa in tension and compression, and an ultimate tensile strength of 0.1–0.3 MPa. So as to situate this report, a *P. ostreatus* mycelium has been reported to exhibit a Young's modulus of up to 28 MPa for an ultimate strength of 0.7 MPa, and a *G. lucidum* mycelium a Young's modulus of up to 12 MPa for an ultimate strength of 1.1 MPa[17]. Beech wood has a Young's modulus of 11.9 GPa at 12% moisture content, and 9.5 GPa when green[33]. Beech wood particles are therefore important load-carrying members of the system and reduce the magnitude of stress experienced by the mycelial matrix. The plastic strain of the composite is contributed to only by particles in such composite, which is clearly exhibited in the range of results in Fig. 4. As introduced with the common reed and rattan containing specimens, the dewetting behaviour of the larger particles or reinforcements present in the composite is a principal contributor to damage nucleation. Furthermore, the shape, nature, and distribution of particles in a two-phase composite has been shown to have a substantial influence over the load transfer between members and hence their overall stiffness[3]. Moreover, while lignin is a primary contributor of strength parallel to grain, hemicellulose supports compression strength perpendicular to grain. Its decay greatly affects the structural integrity of wood and its hardness[34].

The results of the experimental series are plotted as normalised by density in Fig. 8, and on an Ashby map for elastic and density (Fig. 9). In both figu es compressive characterisation from the published MBC state-of-the-art

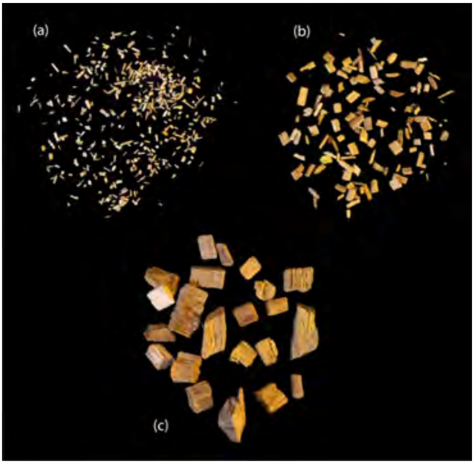

Figure 7. Beech wood particles of small (**a**), medium (**b**) and large (**c**) granulation used in this study.

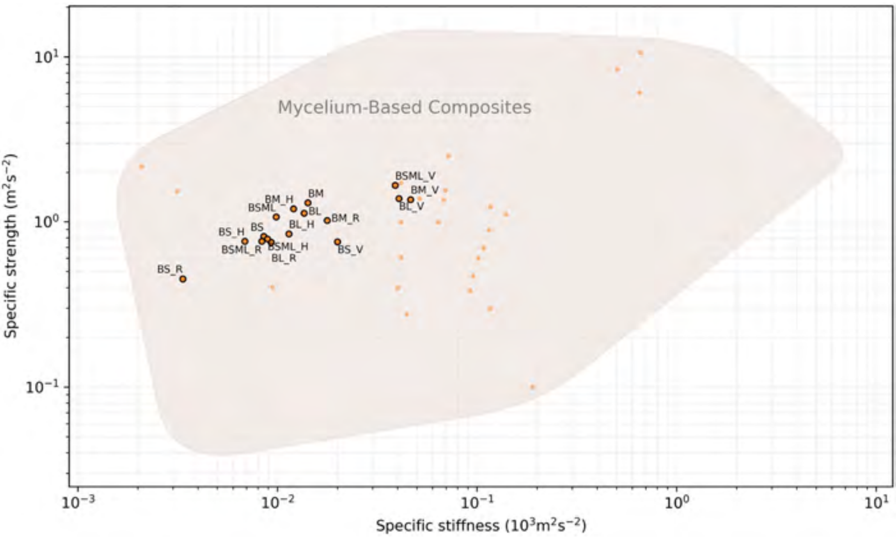

Figure 8. Specific trength results as a function of specific tiffness. Labelled data points: results from this study; unlabelled data points: reports from the state-of-the-art.

are plotted[4–7,35–43]. These figures gather evidences produced with approximately ten fungal species, two studies having not disclosed the ones they used[6,36]. There are 69 data points gathered from thirteen journal and conference articles. These include articles reporting on strength and/or stiffness in compression; 6 data points had no density reported[7]. Only the reports with sufficient data are rendered on the figures.

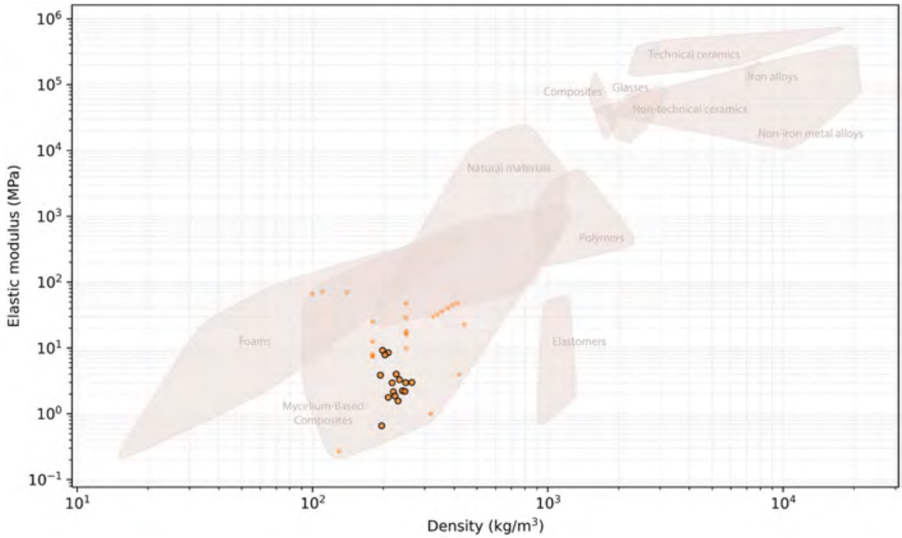

Figure 9. Young's modulus results as a function of density. Circled data points: results from this study; uncircled data points: reports from the state-of-the-art.

While wood particles are of common use in MBC, other substrates have been used such as non woven cotton fibres[13]. A small number of studies have investigated the addition of non-organic aggregates to a lignocellulosic substrate for improving its stiffness, such as with carbonate sand[38], and sand and gravel[44].

Statistical analysis

Six replicates were produced and tested for each of the specimen types, the distributions are two-tailed. The mean of Fisher's defi ed kurtosis for Young's modulus series is − 0.3328 (s.d. 0.9353) and − 1.0564 for ultimate strength (s.d. 0.5343). Fisher–Pearson's skewness coefficient mean for Young's modulus is 0.5834 (s.d. 0.8540), and 0.1733 for ultimate strength (s.d. 0.5137). The distributions are considered normal[45], which was verifi d for ultimate strength and Young's modulus results with the Shapiro–Wilk test (respectively $p = 0.9224$ and $p = 0.0030$, α=0.001). Equality of variances was therefore controlled with the Levene test; Young's modulus result variances are not equal ($p = 1.3940\text{e}{-05}$, α=0.05), neither are ultimate strength ones ($p = 0.0459$, α=0.05). Welch's ANOVA was conducted for the two parameters: fibre placement for Young's modulus and ultimate strength (respectively $p = 0.0001$ and $p = 0.0013$), and particle size for Young's modulus and ultimate strength (respectively $p = 0.0030$ and $p = 4.6462\text{e}{-09}$). The mean values of specimen groups are significantly different (α=0.005). Using the pairwise Games-Howell test we identified the most significant reinforcement to be the fibre coaxial to load against fibre perpendicular to load, the control, and hessian jacketing (all $p = 0.001$ as per Young's modulus; respectively $p = 0.030$, $p = 0.004$, and $p = 0.004$ as per ultimate strength; α=0.05). Continuing this test, we identifi d the most signifi ant aggregate size to be the 0.5–1.0 mm interval (BS family) against the BM and BL families (respectively $p = 0.029$ and $p = 0.036$ as per Young's modulus; all $p = 0.001$ as per ultimate strength; α=0.05). The BS family had a significant difference to the BSML family as per aggregate size over ultimate strength ($p = 0.001$, α=0.05), but not over Young's modulus ($p = 0.106$, α=0.05).

Conclusions

Across the literature, we find that the lack of a unified approach in the use of analytical models and/or methodological approaches has resulted in inconsistency with specimen design, cultivation and testing protocols. Th s raises the question of portability and comparability of results. The adoption of the two-phase particulate composite model helped us identify ASTM D1037 as the most appropriate candidate to support the design of specimens and of the experimental plan. As a general observation, we found that specimens using particles in the 0.75–3.0 mm range resulted in a higher strength and stiffness in compression.

We extended this study to fibre placement strategies with three typologies: rattan fibres perpendicular to load, common reed fibres coaxial to load, and hessian jacketing coaxial to load. The addition of fibre coaxial to load and hessian jacketing had a significant effect over Young's elastic modulus and ultimate strength (α=0.05). Fourier-Transform Infrared (FTIR) spectrometry was used to qualify (1) the materials used as principal substrate and fibre addition, (2) the mycelium of *G. lucidum*, (3) and *G. lucidum* colonised beech wood. We found that the *G. lucidum* species degraded primarily lignin and hemicellulose before cellulose, in accordance with previous observations[10,46].

Because of the wide range in particle sizes used and fibre composition typologies, the significant difference between specimen groups supports our hypothesis that the two-phase particulate model is suited for future MBC studies (α=0.005). These studies might involve exploring a wider variety of particle shapes, natures, and distributions as these parameters have been shown to have a significant influence over the elastic and plastic behaviour of composites[3]. We demonstrated that the modifying of specimens could be attained with contrasting examples of coaxial reinforcement and perpendicular fracture initiators, with signifi ant effect (α=0.005). However, it should be noted that fibre placements were subjected to variability as fibres could partially misalign with the load axis or its perpendicular during production. Th s suggests that the standard deviation of the results can be reduced by improving the accuracy in manufacturing.

Specific strength and stiffness of the results is plotted on Fig. 8, where we can notice the increased composite efficiency for the fibre coaxial to load series. The resulting behaviour of the specimen groups is plotted onto an Ashby map and presented in Fig. 9. The composites display an average performance as compared to the MBC state-of-the-art, and interestingly consolidate the existence of a material pole situating in between foam and elastomer behaviour, as per Young's modulus. Commercial applications for such materials typically situate as EPS or XPS sustainable alternatives for insulation or packaging applications, while the elastic modulus of EPS is 6.5–265 MPa for a yield strength of 0.04–10.9 MPa. For its biochemical profile and impact on phylogenetic mycelial expression, or mechanical interest both at the scale of the particles or engineered artefact, heterogeneous and functionalised substrate design for MBC is a scarcely studied yet promising fi ld of research towards market-ready sustainable and creative applications.

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Author contributions

A.R. and P.A. conceived the experiments, A.R. conceived the methodology, A.R. and P.A. conducted the experiments, A.R. analysed the results. All authors reviewed the manuscript.

Competing interests

The authors declare no competing interests.

Additional information

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D.2 Effect of Composition Strategies on Mycelium-Based Composites Flexural Behaviour.

Article

Effect of composition strategies on Mycelium-Based Composites flexural behaviour.

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Abstract: Mycelium-Based Composites (MBC) are a promising class of relatively novel materials that leverage mycelium colonisation of substrates. Being predicated on biological growth, rather than extraction based material sourcing from the geosphere, MBC are garnering attention as potential alternatives for certain fossil-based materials. In addition, their protocols of production point towards more sustainable and circular practices. MBC remains an emerging practice in both production and analysis of materials, particularly with regard to standardisation and repeatability of protocols. Here, we show a series of flexural tests following ASTM D1037, reporting flexural modulus and flexural modulus of rupture. To increase the mechanical proprieties, we contribute with an approach that follows the composition strategy of reinforcement by considering fibre topology and implementing structural components to the substrate. We explore three models that consist of the integration of inner hessian, hessian jacketing and rattan fibres. This investigation demonstrates that the composition strategies result in a significant strengthening on the flexural modulus, not only confirming that mycelium is responsible for the internal bonding but that fibre orientation and size have the major effect on the flexural mechanical proprieties of MBC.

Keywords: Mycelium-Based Composite; Biomaterials; Natural composites

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1. Introduction

Mycelium-Based Composites (MBC) are a novel field of material development leveraging wood-decaying basidiomycota to bind lignocellulosic particulate media via Solid-State Fermentation (SSF). Since 2006 and the establishment of the first commercial venture for MBC products (Ecovative Design LLC) the development of these materials has been supporting packaging and insulation use cases. Both these cases are relevant with a state-of-the-art that has favoured local replication of the production process across the globe and across institutions before advancing studies of the material mechanical model for assessing alternative use cases. The variety of versatile fungi that can be used, coupled with the extensive variety of lignocellulosic substrates with respect to aggregate geometries and chemical profiles, supports a wide design space. The spread of the MBC state-of-the-art regarding only reported stiffness and strength is a strong testimony to this (Fig.4 and 5), while research has yet to lead to functional poles definition within this space [1]. Furthermore, the principal advantage of MBC designs lies in their higher potential for upcycling agricultural wastes, leading to the production of resource conscious and biodegradable materials with a low environmental impact [2] that can contribute to shifting towards a circular economy.

Across the literature, three principal design strategies for modifying MBC mechanical behaviour have been identified: densification, composition, and supplementation (targeting mycelium properties, based on chemical tuning of the substrate) [1]. Mechanically, MBC have been investigated mainly as per their compressive and flexural behaviour [3–12]. Following a previous investigation of the effect of composing the dispersed phase of the composite and qualifying its compressive behaviour [13], we focus in this study on the effect of composition over the flexural behaviour.

Densification is a strategy for stiffening the composite by increasing the density of the substrate by dense packing, cold or hot pressing. Densified specimens have been reportedly leading to an increase of flexural modulus between 27 and 72-fold, and 4 to 14.5-fold increase in flexural strength for a densification from 100 – 130 kg/m^3 to 350 – 390 kg/m^3 [9]. While staying at the lower end of MBC densities, another study reported an increase of 4.4-fold in flexural strength while increasing composite density by a factor 1.4 (from 102 to 141 kg/m^3) [4]. Composition is a second strategy for modifying MBC behaviour by adding structural components to the substrate, including, for instance, orientated fibres or textiles. Modifying particle properties is also considered an instance of the composition strategy [1]. The MBC state-of-the-art largely considers monolithic and homogeneous composites, besides a few study groups investigating jute type materials in sandwich composite reinforcement, and wood panels introduction [10,14,15]. A hybrid protocol using both composition and densification techniques has been employed by using blend bacterial cellulose (BC) produced by a *Komagataeibacter xylinus* colony. The BC cellulose fibrils were mixed to the hemp fibres serving as principal substrate, and set to be colonised by a *Trametes versicolor* species [6]. This method targets an increase in aggregates binding. BC introduction did not result in a statistically significant difference. Nonetheless, pressing temperature change from 70 °C to 200 °C led to a 1.42-fold increase in stiffness and a factor 1.54 increase in flexural strength to reach 2.94 MPa.

In this study we take agency on the design of the composite and its substrate, following the method of composition as a reinforcement strategy we introduced orientated fibres and hessian. Following ASTM D1037, we report on three point bending for three categories of composition: the embedding of a hessian arming at mid-thickness, hessian jacketing, and rattan arming in specimen length. Apart from the inner hessian group (BM_I), the introduction of rattan fibres and hessian jacketing led to significant increases in both strength and stiffness (α=0.05).

2. Materials and methods

Referenced standards for evaluating flexural properties of MBC in the state-of-the-art are presented in Table 1. ASTM D1037 was used as the standard evaluation method. We use this standard because it is the most referred set of guidelines in MBC development, covers various tests and refers to the fittest material model. We report on three point bending.

To this end, four specimen groups were designed:

- Control: no fibre,
- Inner hessian: a flat layer of hessian was introduced at mid-thickness,
- Hessian jacketing: a hessian jacketing was introduced in the length,
- Rattan: five parallel rattan fibres of 6 mm diameter by 500 mm, separated by 8 mm, were introduced in the length and at mid-thickness.

The wet specimens are parallelepipeds of 520 x 72 mm, with a nominal thickness of 20 mm. The width and thickness were not affected by the desiccation, but the length of the dry specimens varied between reinforcement strategies and shrank on average by 3.5 % in the control group, 2.7 % in the inner hessian group, 1.7 % in the hessian jacketing group, and 0.8 % in the rattan group. Six replicates were produced and tested for each of the specimen types. Load testing was performed on a Mecmesin MultiTest-dV testing bench equipped with a 2500 N load sensor, with a loading speed of 10.0 mm/min. Flexural modulus and modulus of rupture were calculated following ASTM D1037.

Materials

A millet-grown spawn of species *G. lucidum* (reference M9726) was acquired from Mycelia BVBA (Nevele, Belgium). The spawn was stored at a constant 4 ºC and 65 % RH prior to being used. The principal substrate of the specimens is European beech wood (*Fagus sylvatica*) of a 0.75 – 3.0 mm granulation (Räuchergold type HB 750/2000, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany). Longitudinal reinforcement was introduced in the BM_R specimens group by using 6 mm diameter rattan fibres (*Calamus manan*; B.V.

Standard	Designation	Ref.
ASTM C203	Standard Test Methods for Breaking Load and Flexural Properties of Block-Type Thermal Insulation.	[16]
ASTM C393	Standard Test Method for Core Shear Properties of Sandwich Constructions by Beam Flexure	[10]
ASTM D7250	Standard Practice for Determining Sandwich Beam Flexural and Shear Stiffness	[10]
ASTM D1037	Standard Test Methods for Evaluating Properties of Wood-Base Fiber and Particle Panel Materials.	[17–19]
ISO 16978	Wood-based panels – Determination of modulus of elasticity in bending and of bending strength.	[5]
ISO 12344	Thermal insulating products for building applications – Determination of bending behavior.	[5]

Table 1. Referenced standards for flexural characterisation in the MBC state-of-the-art.

INAPO, Bloemendaal, Netherland), and hessian was used for the BM_I and BM_H groups (*Cannabis sativa* subsp. *sativa*; NEMO Hemp jam web 370 g/m^2, Naturellement Chanvre, Echandelys, France).

Cultivation protocol

The principal substrates, fibres and hessian were prepared at 40 % MC with mineralized water and sterilised at 121 °C for 15 min. The principal substrates were then mixed with 16 wt% spawn and incubated in polypropylene filtered bags (SacO2, Deinze, Belgium) for 7 days at 27 °C in the dark. Once colonised, the principal substrates were broken down and formed with the sterile fibre and hessian into alcohol cleaned aerated PETG moulds. The formed specimens were incubated for 21 days at 27 °C in the dark, then oven-dried for 48 hours at 60 °C. The dried specimens were stored at 4 °C and 65 % RH prior to testing. No external mycelium was cultivated on the boundaries of the specimens. No additives were used.

3. Results

We investigated the effect of diverse reinforcements on the mechanical behaviour in flexion of MBC using three levels: inner hessian (BM_I), hessian jacketing (BM_H), and rattan fibres in the length (BM_R). These strategies are illustrated in Fig.2. Figure 3 shows the specimens after testing. Isotropic controls were added to the experimental series (BM). Experimental parameters per specimen type and resulting mean density, mean flexural modulus and mean modulus of rupture are presented in Table 2. Box plots of the results for flexural modulus and modulus of rupture are presented in Fig.1. It can be noted that the mechanical failure of the specimens was related to dewetting of the principal substrate. Rattan fibres did not fail nor deform plastically. Likewise, no debonding of the hessian jacketing was visible after testing. In Fig.3 we can observe some of the tested specimens. It is noted that the principal substrate phases have all been softened but remain integrated with no visible external fractures.
Similarly to our previous investigation on compression behaviour characterisation [13], the hessian jacketing series offers a contrasting instance of the effect that the cultivation of an external mycelial skin on test specimen might lead to. The BM_H series resulted in a 1.95-fold increase in flexural modulus as compared to the control group, and 1.5-fold increase in strength. In contrast to the BM_I series which had a hessian reinforcement added at mid-thickness, jacketing is a particularly suitable fibre composition strategy as the

low elasticity of hessian contributes to resisting the tensile stress on the surface opposite to the load and on covered side surfaces. The BM_R series has the highest relative standard deviation with 41.3 % of the mean, while other groups have a standard deviation of a maximum of 27.2 %. An extremum is reported at 2.44 GPa while Q3 is 1.35 GPa for a mean at 1.34 GPa. If not considering this extremum, the mean of the series is at 1.12 GPa for a standard deviation closer to MBC standards at 211.06 MPa (18.8 %).
The results of the experimental series are plotted on an Ashby map (Fig.5) and are presented as normalised by density (Fig.4). In the latter figure we can notice the increased mechanical efficiency of the BM_R series thanks to the composition strategy. In both figures flexural reports from the published MBC state-of-the-art are plotted. These two figures gather evidences produced with approximately ten fungal species, two studies having not disclosed the ones they used [8,10]. There are 42 data points gathered from ten journal and conference articles. These include articles reporting on strength and/or stiffness in flexion; 3 data points had no density reported [12]. Only the reports with sufficient data are rendered on the figures.

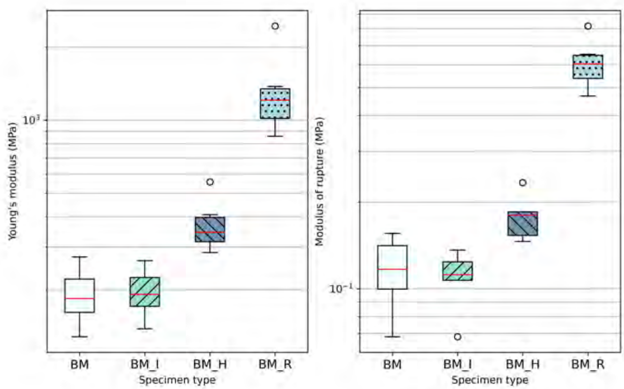

Figure 1. Flexural modulus (a) and modulus of rupture (b) box plots results.

Statistical analysis

The result distributions are two-tailed. The mean of Fisher's defined kurtosis for flexural modulus series is −0.7243 (s.d. 0.6040) and −0.5844 for modulus of rupture (s.d. 0.5019). Fisher-Pearson's skewness coefficient mean for flexural modulus is 0.5582 (s.d. 0.4907), and −0.1625 for modulus of rupture (s.d. 0.7697). The distributions are considered normal [20], but did not satisfy the Shapiro-Wilk test (modulus: $p=1.1266e-5$, modulus of rupture: $p=3.4568e-5$, $\alpha=0.001$). Equality of variances was controlled with the Levene test; flexural modulus result variances are equal ($p=0.0208$, $\alpha=0.05$), such as modulus of rupture ones ($p=0.0146$, $\alpha=0.05$). One-way ANOVA was conducted for flexural modulus and modulus of rupture regarding reinforcement strategies (respectively $p=1.8619e-6$ and $p=2.8678e-11$). The mean values are significantly different ($\alpha=0.001$). Using the pairwise Games-Howell test we confirm the significant difference between the rattan group and the other groups for flexural modulus and modulus of rupture ($\alpha=0.05$). The inner hessian group was not significantly different from the control ($p=0.900$).

Type	**Fibre composition**	**Mean density** (s.d.)	**Mean flexural modulus** (s.d.)	**Mean modulus of rupture** (s.d.)
BM	Control	232.24 kg/m^3 (18.24)	192.71 MPa (52.40)	0.12 MPa (0.03)
BM_I	Inner hessian	227.22 kg/m^3 (8.46)	197.33 MPa (45.56)	0.11 MPa (0.02)
BM_H	Hessian jacketing	236.75 kg/m^3 (12.00)	375.14 MPa (98.81)	0.18 MPa (0.03)
BM_R	Rattan	249.48 kg/m^3 (9.78)	1.34e3 MPa (570.68)	0.61 MPa (0.12)

Table 2. Summary of specimen types parameters, resulting dried densities, and flexural properties.

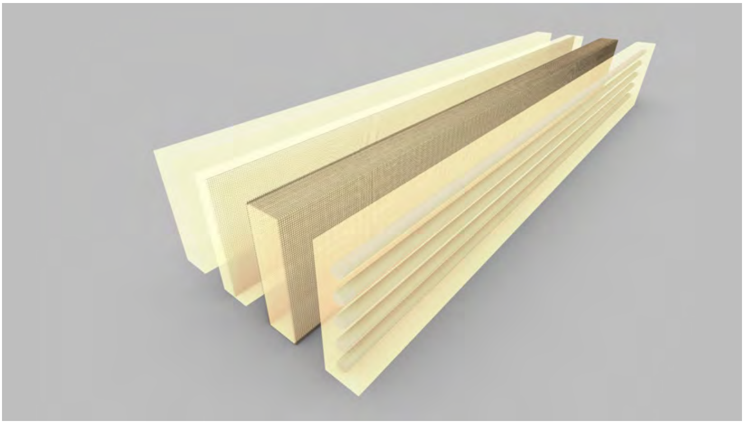

Figure 2. Fibre placement strategies (left to right): control (BM), inner hessian (BM_I), hessian jacketing (BM_H), rattan fibres (BM_R).

Figure 3. Fibre placement strategies in tested specimens (left to right): rattan fibres (BM_R), hessian jacketing (BM_H), inner hessian (BM_I), control (BM).

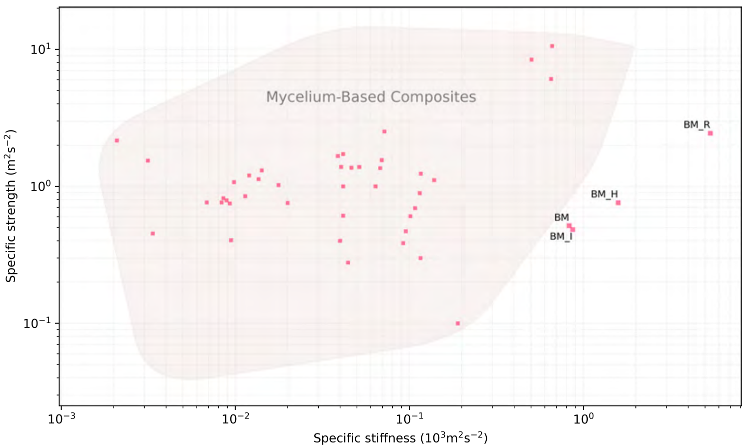

Figure 4. Specific strength results as a function of specific stiffness. Labelled data points: results from this study; unlabelled data points: reports from the state-of-the-art.

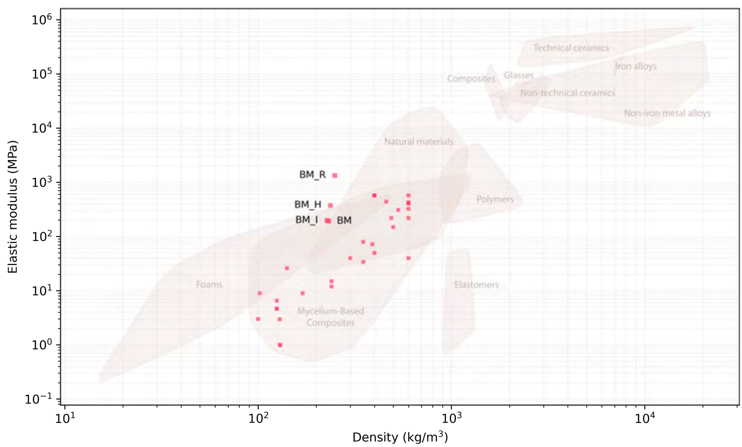

Figure 5. Flexural modulus results as a function of density. Labelled data points: results from this study; unlabelled data points: reports from the state-of-the-art.

4. Conclusions

The investigation presented in this paper has focused on the use of natural fibre composition in MBC as a means to modify flexural behaviour. We have demonstrated three fibre composition designs and show significant increases in stiffness and strength comparing the BM_R and BM_H series to the BM control group (α=0.05). The mean of the flexural modulus for the most performative BM_R series (1.34 GPa) is still close to three times lower than that of Medium-Density Fibreboard (MDF; 4 GPa), and approximately 16 times lower in modulus of rupture (0.62 MPa, MDF: 10 MPa). Considering the higher density of MDF (750 kg/m^3) as compared to the BM_R series (249.48 kg/m^3) a future investigation could focus on a hybrid strategy of composition and densification so as to improve aggregate interlocking and resulting strength and stiffness. The arming strategy adopted with the introduction of inner hessian did not prove to be significant. We reflect that as it was positioned at mid-height of the specimens it was theoretically not subjected to tension and could only partially contribute to the bending stiffness. Future investigations may focus on various soft arming positioning strategies reflecting on the most mechanically demanding areas of the specimens as per a defined use case. Composite production accuracy improvements are expected to contribute to reducing the standard deviation of results. Furthermore, reinforcements may be strategised in developing efficient or multi-functional composites, for instance, in designing the principal substrate to be thermally performative in addition to introducing reinforcements to perform structurally. We also report on specimens dimensional stability after drying linked to the diverse compositions, this aspect can be investigated in the future both as a means to explore design consequences, and for production control.

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E Book section

E.1 Design Strategies for Mycelium-Based Composites

Design Strategies for Mycelium-Based Composites

20

Adrien Rigobello and Phil Ayres

Abstract

Mycelium-based composites (MBC) can be designed utilising a wide range of lignocellulosic substrates and widely distributed versatile ligninolytic fungi. While a wide range of mechanical behaviour has already been reported in the past 15 years, showing potential to obtain viable products for a variety of uses; no systematic description of the engineering parameters has been established till date. We review carbohydrate-active enzyme (CAZyme) activities of fungal species, lignocellulosic substrate chemical profile at cellular level, wetting characteristics, substrate aggregate and composition characteristics. We identify three principal strategies for designing MBC: supplementation, densification and composition, and discuss them regarding outstanding reports from the state-of-the-art. We report on solid-state fermentation supplements having significant effect on fungal CAZymes activities (e.g. monosaccharide, nitrogen, ash, pH of buffer). State-of-the-art designs and process control promote specific enzymatic activities independent of species genomics; systematically investigating supplementation, densification and composition design strategies in the future may lead to both a widening and deepening of the available material qualities, along with a focus on developments around functional poles. Additionally, future reproducibility studies of MBC development reports may both improve the overall market readiness and public adoption of MBC solutions and valorise the wealth of material and design semiotic properties that the versatility and affordability of MBC systems support.

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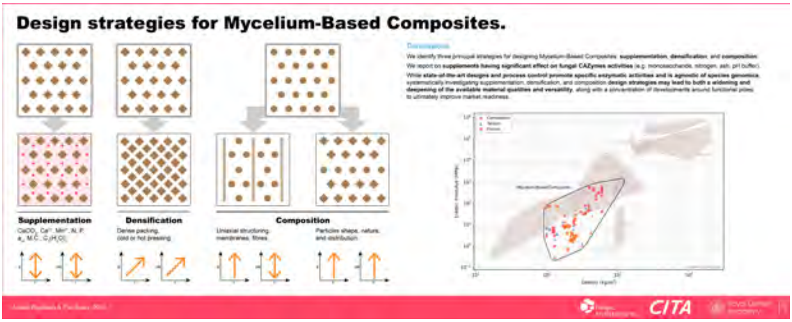
Graphical Abstract



Keywords

Solid-state fermentation · Ligninolytic fungi · Mycelium-based composites · CAZymes · Biocomposites · Biomaterials

Highlights

- We identify three principal strategies for designing mycelium-based composites: supplementation, densification and composition.
- We report on supplements and wetting metrics having significant effect on fungal CAZyme activities and mycelia phenotypic expression.
- We report on the composition strategies having significant effect on mechanical response of mycelium-based composites.
- We present state-of-the-art mechanical reports comparatively from species and substrate specifics.
- Systematic investigations in future may lead to widening and deepening available material qualities along with a focus on functional poles.

20.1 Introduction

Inherited from the nineteenth- to early twentieth-century method of fungal strain transfer by lignocellulosic solid-state fermentation (SSF) [1], mycelium-based composite (MBC) cultivation protocols can be based on a wide range of substrates containing lignocellulosic polymers using versatile ligninolytic fungi. Widely available basidiomycota have been instrumental in developing composites with a wide range of mechanical performance, showing potential to obtain viable products for a variety of uses. However, no systematic description of the engineering parameters has been established till date. Although the first MBC commercial venture was established in 2006 (Ecovative Design LLC), an aggressive patent strategy combined with an inertia in explaining the engineering-driving principles of MBC has resulted in a very low market readiness and market permeability of the material solutions. With a global urgent need to adopt low-energy and low-impact alternatives to existing detrimental material systems on a scale, we argue that rationalising and systematising approaches to analysing MBC systems is necessary to actualise their potential and facilitate market readiness.

As composites, MBC are made of a matrix phase that corresponds to a continuous and foam-like or elastomer-like mycelium, and a dispersed phase composed of particles and/or fibres. Additionally, surface or volumetric components can be used to compose the substrate of a composite and modify its properties. Fungal decay is functionalised as a binding method between elements of the dispersed phase. From this generic definition, we can appreciate the sheer breadth of the material design space, and can identify the main material characteristics for defining the cultivation protocol and specifying composite properties:

- Enzyme arrays of fungal species,
- Substrate chemical profile,
- Substrate wetting characteristics,
- Substrate aggregate characteristics,
- Substrate density and composition.

The resulting composite properties depend on the SSF model that is adopted. While tacit knowledge of MBC craft is sufficient for reaching a satisfactory output, thanks to the competitive colonisation dynamics of the fungi in use, the explanation and modelling of the driving parameters of MBC systems can foster a deeper engagement through engineering practice, thus extending the reach and effectiveness of material research and improving market readiness. Paste-like substrate design for fungal colonisation is not studied at present. The inner structure of a typical MBC is presented in Fig. 20.1.

We start by presenting a review of the MBC model from a ligninolytic fungi enzymatic activities perspective, and proceed to describe the biochemistry of

Fig. 20.1 Typical inner structure of an MBC

lignocellulosic substrates while correlating it to fungal enzymatic abilities. Building upon studies of fungal niches in the wild and their parasitic or saprotrophic behaviour in trees, we come to understand the hydrolytic abilities of wood in relation to their chemical composition and evolutionary stage as a primary parameter for predicting fungal colonisation. A comprehensive review of supplementation methods for influencing enzymatic activities is presented for ultimately adapting protocols to locally available resources. Furthermore, in describing the behaviour of MBC from this perspective, we review studies that have shown a clear correlation between the mycelial mechanical properties and their cultivation substrate chemical composition. While these properties have been demonstrated to be modifiable to result in foam-like or elastomer-like behaviour, nature of substrate aggregates (size, distribution and shape) are the principal contributors of composite strength and stiffness. Outstanding mechanical results from the MBC state-of-the-art are then discussed with regard to these insights, in an effort to describe the behaviour of composites. We identify three main substrate-based exploration strategies for the MBC system: densification (by dense packing, cold- or hot-pressing), composition (by introducing structuring elements) and supplementation (targeting mycelium properties, and based on chemical tuning of the substrate). Furthermore, we have analysed available data in published peer-reviewed studies to offer a critical review of the state-of-the-art, and conclude on the most significant engineering parameters and recommend research directions.

20.2 Enzymatic Activities of Ligninolytic Fungi

The species of *Trametes*, *Ganoderma* and *Pleurotus* are among the most frequently cited fungal families in MBC design [2]. *Schizophyllum commune* is a less investigated species but there is a continued interest [3], especially because it is one of the few mushroom-forming fungi for which genes have been inactivated by homologous recombination [4]. This strategy has proved useful for genetic studies and functional genomics [5]. Gene inactivation articulates the principles of homologous recombination for replacing an existing autologous gene with a designed heterologous gene. Furthermore, the recombinant DNA constructs of *S. commune* has been reported to express in other mushroom-forming fungi, such as *Pycnoporus cinnabarinus*, which supports its importance for fungal biology [4, 6]. *Irpex lacteus* has been used previously in MBC development [7], as in *Agaricus bisporus* [8] and *Fomes fomentarius* [9]. Other less investigated species have also been reported [2].

Wood-rotting fungi can be classified by their decay activity, which cover three main modes: white-rot (specified as selective delignification and simultaneous rot), brown-rot and soft-rot. Across the surveyed species, all of them are white-rot, ligninolytic fungi. They have the enzymatic ability to decompose all three principal polymer classes found in wood: lignin, hemicellulose and cellulose, but most often degrade primarily lignin and hemicellulose as reported in *Ganoderma lucidum* [10]. In such cases, these species qualify as selective delignifies, as opposed to simultaneous decomposers where all three polymer classes endure similar decay rates. Brown-rot fungi use a two-step oxidative–enzymatic mechanism for the breakdown of cellulose and hemicellulose [11]. Their lignin degradation is limited to modification in lignin, although brown-rot fungi is a polyphyletic group evolved from at least seven white-rot lineages [12]; they reportedly lack more than 60% of the genes known to be present in white-rot species. Brown-rot species degrade cellulose and hemicellulose at a higher rate than white-rot species in laboratory studies [13]. This decay mode is most often encountered in coniferous trees. Soft-rot fungi commonly share the ability to demethylate. This mode is associated with Ascomycota and monokaryotic mycelia, and is distinctive for its tunnelling decay pattern through lignin layers to reach cellulose and hemicellulose-rich areas [14]. Lignicolous fungi can benefit from a variety of enzymatic strategies to ultimately fully decompose lignocellulosic substrates. The environmental conditions for enzyme activation define the species niche and their subsequent distribution. The principal wood-rot enzyme families are presented in Table 20.1.

Lignin linkages result from radical reactions, in contrast to polysaccharide polymerisation resulting from hydrogen bonding. Lignin, therefore, does not involve hydrolytic enzymes and cannot be used as a source of carbon or energy for most wood-rot fungi. Its decomposition serves primarily for the fungus to gain access to hemicellulose, pectin and cellulose compounds. Lignin depolymerisation is mostly initiated by aromatic ring oxidation by peroxidases (by use of H_2O_2 or R–OOH) [35], while other aromatic compounds in the cell structure are mostly catabolised by monooxygenases and dioxygenases. Peroxidases are heme proteins, such as lignin peroxidase (LiP) and manganese peroxidase (MnP). LiPs are known to oxidise phenolic aromatic substrates along with various nonphenolic lignin compounds, and other organic compounds with a high redox potential. MnP leads to H_2O_2 oxidising Mn^{2+} to Mn^{3+}, an oxidising compound to monomeric phenols. Mn^{3+} chelates permeate through wood cells and decay lignin selectively. These chelates cannot oxidise nonphenolic compounds, but radicals formed by Mn^{3+} oxidation can, in turn, oxidise benzyl alcohols and other diarylpropane structures. MnP can also peroxidise lipids [36]. Versatile peroxidase (VP), in addition to MnP and LiP, are enzymes that are Mn^{2+} specific, but that can oxidise phenolic and nonphenolic compounds in its absence (as with LiP). They are involved in oxidation of high redox potential aromatic substrates, thus reducing Mn^{2+}-independent oxidation but not affecting Mn^{2+}-dependent one [15]. Mn^{2+}-independent oxidation capabilities for phenols, small dye compounds and amines has also been described for MnP [35], and it is predicted from sequencing that similar divergent MnP may be present in *Trametes* spp., *Pleurotus* spp. and *A. bisporus* among other species [35, 37]. Hydrogen peroxide, necessary for LiP and MnP activities, is the product of fungal enzymatic activity. A peroxidase family with wide substrate specificity, active at pH of approximately 3, has been identified in fungi: dye-oxidising peroxidase (DyP). In addition to oxidising peroxidase substrates, it can perform on anthraquinones. Laccases are a very versatile family of enzymes found in nearly all fungi including those without lignin degrading abilities [38]. These enzymes perform functions ranging from synthesis of melanin and other pigments to conidia and fruiting body formation, and lignin decomposition [39]. These belong to a multicopper enzyme

Table 20.1 Extracellular enzymes in lignicolous fungi and their optimal activation temperature and medium pH [15–34]

Target polymer	Enzyme family	Reaction	Substrate	Temperature (°C)	pH
Lignin	Lignin peroxidase	Oxidation	Phenol, nonphenolic aromatics	28–40	3.0–4.5
	Manganese peroxidase	Oxidation	Manganese ions, phenol, small dye compounds, amines	23–40	5.0–7.0
	Versatile peroxidase	Oxidation	Manganese ions, phenol, nonphenolic aromatics	35	7.0
	Dye-oxidising peroxidase	Oxidation	High redox-potential dyes, nonphenolic lignin model compounds	28–70	3.0–7.0
	Laccase	Oxidation	Phenol, diphenol, aminophenol, aromatic compounds	35–50	4.0–5.0
Hemicellulose	Xyloglucanase	Hydrolysis	β-1,3-Glucans, carboxymethyl cellulose, xyloglucan oligosaccharides	37–75	3.0–9.4
	Xylanase	Hydrolysis	Glycosic bonds in xylan backbone	28–50	3.0–9.0
	Mannan-degrading	Hydrolysis	Mannan, galactomannan	45–92	2.4–7.5
	Polygalacturonidase	Hydrolysis	Pectin	–	–
	Pectin lyase	β-Elimination	Heavily methyl-esterified	50	8.0
	Pectate lyase	β-Elimination	Lower esterification and Ca^{2+}	30–90	5.0–11.5
	Cellobiose dehydrogenase	Oxidation	Cellobiose, cellodextrins, lactose	60	3.0–10.0
Cellulose	Cellobiohydrolase I	Hydrolysis	Crystalline cellulose	40–80	–
	Cellobiohydrolase II	Hydrolysis	Amorphous cellulose, β-xylan	50–63	4.0–6.0
	GH5 endoglucanase	Hydrolysis	Cellulose	45–65	4.5–7.0
	GH7 endoglucanase	Hydrolysis	Crystalline cellulose	45–65	4.5–7.0
	GH12 endoglucanase	Hydrolysis	Amorphous cellulose	45–65	4.5–7.0
	GH45 endoglucanase	Hydrolysis	Crystalline cellulose	45–65	7.0
	GH1 β-glucosidase	Hydrolysis	β-Linked oligosaccharides, aglycone-linked β-glucosides	25–50	5.0–7.0
	GH3 β-glucosidase	Hydrolysis	β-Glucan	25–50	5.0–7.0

family and catalyse the removal of an electron from phenolic hydroxylic groups to form phenoxy radicals, which then polymerise via radical coupling. Ultimately, the reactions lead to the presence of oxidised quinones and couples oligomeric products. Laccases also have wide substrate specificity and can act upon an extended range of phenols, diphenols, aminophenols and aromatic compounds. When their catalytic reaction is accompanied by demethylation, it may lead to ring cleavage. Tying lignin decomposition to cellulose and hemicellulose decay, cellobiose dehydrogenase (CDH) has the ability to oxidise cellobiose, cellodextrins and lactose [16]. Hemicellulose-degrading enzymes are capable of hydrolysing various polymers, β-1,4-galactosidase degrades xyloglucan, xylan, galactomannan and pectin [17]. More specific xyloglucanases, xylanases, mannan-degrading enzymes and pectinases can be found in all fungi to a varied extent. Typically, pectin can be hydrolysed by polygalacturonidases, but also cleaved by β-elimination by pectin and pectate lyases. Cellulose hydrolysis by fungal enzymes employs glycohydrolases (GH) by glycosic bond cleavage. Among GH families, cellobiohydrolase I (CBH I) is the major cellulase being produced in white- and soft-rots; it binds to crystalline cellulose and hydrolyses it into cellobiose. Cellobiohydrolase II (CBH II), found in all but brown-rot fungi, binds to amorphous cellulose and generates cellobiose from the non-reducing end of cellulose using acid catalysis of the β-1,4-glycosic bond. Endoglucanases are a major enzyme group capable of hydrolysing cellulose polymer from within the molecule rather than its end. The variety encountered in fungi refers to four GH families: GH5, GH7, GH12 and GH45. GH5 endoglucanase is a less studied family in fungi. GH7 endoglucanase displays a similar mechanism to CBH I, with the difference that it can hydrolyse from the middle of cellulose molecules. GH12 endoglucanases lack a carbohydrate-binding module (CBM). They are therefore unable to degrade crystalline cellulose but can hydrolyse it in its amorphous form. GH45 endoglucanase benefits from a CBM. β-Glucosidases, produced by all fungi, belong to GH1 and GH3 families. GH1 β-glucosidases are able to cleave soluble β-linked oligosaccharides from up to nine glucose residue chains and aglycone-linked β-glucosides, and can be competitively inhibited by the products of CDH activity. GH3 β-glucosidases can remove single glucosyl residues from the non-reducing ends of oligo- and polysaccharides such as β-D-glucans, β-1,3-D-glucans, β-1,4-D-glucans [40]. Lytic polysaccharide monooxygenases (LPMO) operating by oxidative cleavage have been shown to enhance cellulose degradation [41, 42].

Carbohydrate-active enzymes (CAZymes) have been classified into six principal families: auxiliary activities (AA), glycoside hydrolase (GH), glycosyltransferase (GT), polysaccharide lyase (PL), carbohydrate esterase (CE) and carbohydrate-binding module (CBM). We have summarised the genome distribution of *Trametes versicolor*, *Pleurotus ostreatus* PC15 and *G. lucidum* G0119 according to this nomenclature in Table 20.2 to exemplify specific species decaying modes. *G. lucidum* benefits from the widest known CAZymes array with 565 genes having been assigned to these functions [10, 44]. This is a well-known versatile fungus capable of selective delignification, as we have previously confirmed experimentally [45]. The actual decay strategies in selective delignification-capable fungi has been

Table 20.2 Genome distribution of CAZymes from selected species: *T. versicolor* (TV) [43], *P. ostreatus* PC15 (PO) [43] and *G. lucidum* G0119 (GL) [44]

CAZyme family	Enzymes	TV	PO	GL
AA	Laccase, LiP, MnP, VP, DyP, CDH, LPMO	89	114	96
CBM	GH5-GH7-GH45 endoglucanase, CBH I	47	82	33
GH	GH12 endoglucanase, β-glucosidase, xyloglucanase, xylanase, polygalacturonidase, mannan-degrading, CBH II	222	235	262
GT	Glycosyltransferases	85	67	72
PL	Pectin lyase, pectate lyase	9	23	11
CE	Carbohydrate esterases	19	28	91

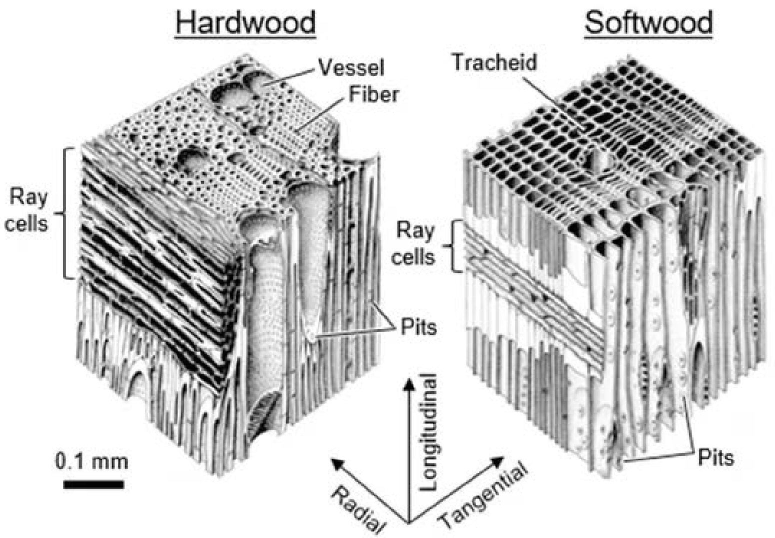
correlated to wood species, lignin contents, temperature, pH and moisture content [46–50].

20.3 Lignocellulosic Substrates

Lignin is the most effective barrier to fungal decay as it prevents migration of low molecular weight diffusible agents (i.e. H_2O_2, veratryl alcohol, oxalate, Fe^{2+}, Fe^{3+}, Mn^{2+}, Mn^{3+}) for decomposing cellulose and hemicellulose. The nature of lignins is of considerable influence on the effectiveness of enzymatic activity. Lignins are polymers of phenylpropene units and they comprise guaiacyl, syringyl and *p*-hydroxyphenyl units. Gymnospermous woods (conifers) are almost exclusively composed of guaiacyl monomers, while angiospermous trees (hardwood) contain approximately equal shares of guaiacyl and syringyl monomers [51]. Similarly, different distributions of hemicelluloses are found in gymnospermous trees (galactoglucomannan, glucomannan and arabinoglucuronxylan are the principal ones) and in angiospermous trees (arabinogalactan, xyloglucan and various glucans are the principal compounds) [14]. Pectins are primarily made of β-1,4-D-galacturonase acid units and their methylesters, interrupted locally by 1,2-linked L-rhamnose units [52]. The primary constituent of wood is cellulose, which is made of repeating glucose units joined by β-1,4-glycosyl linkages. Cellulose represents about 50% of the wood cell wall, lignin 25%, hemicellulose 20–25% and pectin 1–4%. Hemp is often used in MBC practice. Their lignocellulosic components ratio is C:HC:L, which is close to 15:2:1, while their shives display a ratio closer to 2:1:1. The cultivation period and weather has been reported to cause differences in non-cellulosic composition [53].

The variety of wood structures and their chemical composition pairs with various decay strategies driven from the set of fungal CAZyme expression. These wood specifics can be understood from the evolutionary history of cell specialisation. Principal wood structures are presented in Fig. 20.2, reprinted [54]. Wood cells can be made of various specialised cells: tracheids, xylem ray parenchyma, axial parenchyma and vessels. Tracheids are found down to a first evolutionary stage of trees in gymnospermous woods, where they combine the tasks of water transport and

Fig. 20.2 Principal structures of soft- and hardwood; reprinted [54]



structural stiffness. In contrast, gymnospermous woods typically lack vessels and rarely have axial parenchyma. Their tracheid-to-xylem ray parenchyma ratio is most often in the vicinity of 9:1. They contain principally guaiacyl lignin, a product of coniferyl alcohol [55]. In more recent angiospermous woods, water is transported by vessels while tracheids perform structurally. Another evolutionary stage of water transport system in trees can be exemplified by European beech (*Fagus sylvatica*) and birch (*Betula pendula*). Their xylem includes vessels along with tracheids, the former being the only distinct location of guaiacyl lignin. Tracheids and parenchyma contain a fraction of syringyl lignin spread across the cell wall layers, making this secondary evolutionary wood structure attractive for white-rot fungi [14]. A third evolutionary stage can be illustrated by European oak (*Quercus robur*), for which the xylem is ring-porous, containing early and late wood vessels in bands within a thin-walled tracheid matrix. The strength of the wood comes from libriform fibres containing higher syringyl lignin, while tracheids have a higher guaiacyl content [56]. A late evolutionary stage is exemplified by sycamore (*Acer pseudoplanatus*) and ash (*Fraxinus* spp.), where syringyl dominates [57]. Here, vessels are dedicated to water transport, and are jacketed in living libriform fibres that are included amongst dead ones. Guaiacyl lignin has been reported to have a higher resistance to fungal decay [14], which has been corroborated by phylogenetic and experimental studies, showing that catalytic tryptophan presence in more recently evolved peroxidases (located in LiP and VP families) is more efficient at oxidising angio-spermous woods, while others perform better in gymnospermous woods [58]. Early-

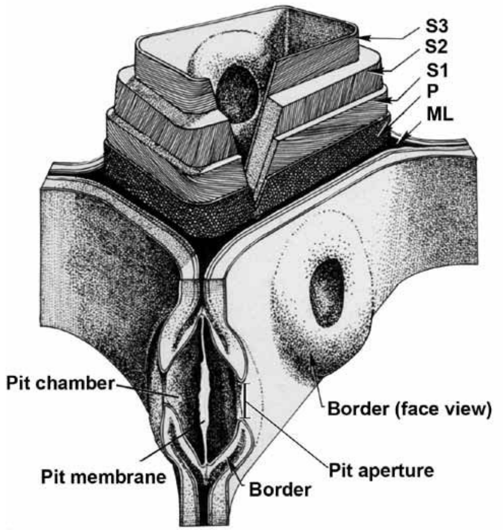


Fig. 20.3 Wood cell wall structure: middle lamella (ML), primary wall (P), secondary wall (S1, S2, S3); reprinted [59]

stage parenchyma decomposition, and a resistance from libriform fibres to white-rot decay, has been confirmed experimentally [14].

The structure of the wood cell wall composing the xylem divides between the middle lamella (ML), primary wall (P) and three layers of secondary wall (S1, S2, S3). They are represented with their fibre orientation and indicative thicknesses in Fig. 20.3, reprinted [59]. The structuring middle lamella is composed of lignin, calcium and pectic compounds that are amorphous. Crystalline cellulose is most prominent in younger wood, so it dominates the S3, and is found in lowering percentages moving towards the P. Inversely, hemicellulose can be found to a greater extent in the P and in reducing shares towards the S3, while lignin is quite evenly spread across the cell wall, with higher concentrations in the ML. The distribution of lignocellulosic compounds in woods is presented in Table 20.3. Colonisation happens through pit apertures present in wood, or the inducing of boreholes by enzymatic activity. Although pectin contents are marginal in wood, pectinases expression is critical to decomposing the pit membrane (Fig. 20.3), which also contains cellulose and allows the fungus to proceed in colonising [61]. Calcium is associated with pectin in wood; Ca^{2+} can combine with fungal oxalic acid and result in pectic acid which solubilises pectin [52]. Calcium, potassium, phosphorus and magnesium are the major elements associated with plant fibre. The mineral content (ash) in softwood is 0.02–1 wt% and 0–5 wt% in hardwood [62]. We can note that large concentrations of calcium oxalate and residues of MnO_2 accumulate during white-rot decay [50]. Moreover, wood can contain a variety of toxic

Table 20.3 Approximated lignin distribution in the different layers of the wood cell wall; adapted [60]

Wood type	Cell type	Layer	Share of total lignin content (%)	Lignin concentration (%)
Gymnospermous early wood	Tracheid	S1–S3	65	24
		ML, P	21	49
		Cell corners	14	64
Gymnospermous late wood	Tracheid	S1–S3	75	22
		ML, P	14	51
		Cell corners	11	78
Angiospermous wood	Fibre	S1–S2	60	19
	Fibre	ML, P	9	40
	Fibre	Cell corners	9	85
	Vessel	S1–S3	9	25
	Vessel	ML, P	2	40
	Ray cells	S1–S3	11	25

compounds, mainly polyphenols or tannins in angiospermous trees and phenolic compounds in gymnosperms. Selective delignification-capable fungi can degrade polyphenols. *Ganoderma* spp. are able to degrade polyphenolic compounds and show chemotropic growth towards these [63]. Nitrogen availability is critical to fungi for enzyme synthesis; the limited amounts in wood makes it resilient to decay (the C:N ratio of birch wood is typically of 55 and 401 in sycamore wood). This compound is mostly found in parenchyma and is known to migrate towards the bark surface during log desiccation [64]. Moreover, wood cell walls specialise depending on local plant mechanical stress and taxonomic order (Fig. 20.4, reprinted [65]), which may lead to a more specific consideration of lignocellulosic substrates to establish cultivation protocols.

The wide distribution of wood-decaying fungi suggests, with increasing evidence, that local environmental factors and substrate specifics drive and regulate colony extent between the various fungal species present. Furthermore, the pH of wood is most often in the 4.0–5.5 range (beech: 5.11, birch: 5.29, for instance) [66]. It has been demonstrated that substrate pH is also influenced by the action of decay [46]. It has been reported, by comparative transcriptonics, that white-rot related genes of a *Phanerochaete carnosa* species would all express in fir (*Abies balsamea*), pine (*Pinus contorta*), spruce (*Picea glauca*) and maple (*Acer saccharum*), but to a varied extent. For instance, MnP genes read was 2.42 times higher in fir compared to maple, but LiP-related genes read was 4.7-fold higher in

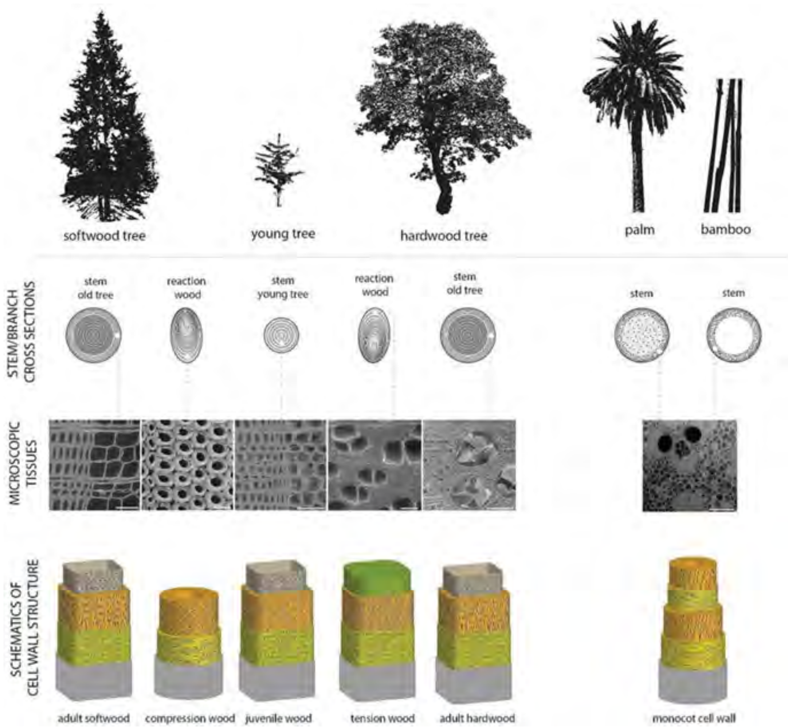


Fig. 20.4 Structure of arboreal plants; reprinted [65]. Microscopic tissues from left to right: adult spruce wood (bar: 50 µm), spruce compression wood (bar: 20 µm), juvenile spruce wood (bar: 50 µm), poplar tension wood (bar: 50 µm), adult oak wood (bar: 200 µm) and vascular bundle of bamboo (bar: 200 µm). Last row showing schematics of cell wall structures. Light grey indicates the compound middle lamella (0.5–1.5 µm) and primary cell wall (approximately 0.1 µm); yellow: S1 layer with a thickness of 0.1–0.35 µm; orange: S2 layer as 1–10 µm thick; inner light grey layer shows the 0.5–1.1 µm thin S3 layer; the green G-layer in tension wood fibres can fill the whole lumen and the cellulose fibrils are oriented along the longitudinal cell axis

maple [67]. This indicates both the resilience of species to the use of a variety of vernacular substrates, but also a precariousness considering the sheer distribution of competitors of an individual species. This can lead to fungi chemically wrestling for resource [68]. *Ganoderma* spp. have been recognised as having the competitive advantage of being able to degrade reaction zones in trees [63]. Studies of lignicolous fungal ecological distribution point to water content and temperature variations being the principal drivers of selection [69]. Climate change has then been related to shifts in fungal niches across hosts [70]. In the wild, host tree species and their evolutionary stage can have an influence on patterns of decay; the tree group that has the most diffuse xylem vessel system may result in a slower initial colonisation. Similarly, gymnospermous wood having 3–4 mm long tracheids benefits from a cellular compartmentalisation strategy to reduce fungal decay, despite its guaiacyl lignin content. These topological effects are prominent at the early stage of fungal colonisation [64].

20.4 Water in Fungal Decay

The presence of capillary water in the substrate is a prerequisite to allow extracellular transport of metabolites in fungi, and to establish osmotic potential for allowing turgor pressure and subsequent hyphal growth. Turgor also enables fungi to penetrate solid substrates [71]. It has been reported that the radial growth of white-rot species *Physisporinus vitreus* at pH 5 and 25 °C increased from just below 0.5 mm/day at $a_w = 0.97$ to 4.5 mm/day at $a_w = 0.998$ [72]. The production of enzymes by fungi and their catalytic activity has been related to water activity with varying local optima depending on the depressor that was used in the study, pointing to $a_w \geq 0.99$ for optimal catalytic activity here [73].

SSF requiring the absence of solution in between particles for a mycelium to develop, the substrate water holding capacity is an important parameter for ensuring an optimal fungal decay and growth [11, 73, 74]. In timber, moisture can exist as bound water within cell walls below the fibre saturation point (FSP), free liquid water in cell cavities above FSP and water vapour. Relative humidity (RH) of up to 97–98% is generally considered to correspond to cell wall uptake (hygroscopic range), and as RH moves towards saturation, wood cell void uptake tends to become more significant in the moisture distribution (over-hygroscopic range) [75]. The latter is conventionally considered as an optimal range for optimal fungal growth in lignocellulosic SSF. The validity of this convention is nonetheless discussed as it was shown that *T. versicolor* would colonise European beech wood at a minimum moisture content (MC) of 15% for a mass loss over 2% (equivalent to 75–80% RH at −1–37 °C); [76]. Brown-rot fungi grow at 92–97% RH, and that instrumental limitations are encountered in the over-hygroscopic range, thus complicating measures and experimental control [75]. For instance, the FSP of European beech wood has previously been determined to be 24.02% MC at 97% RH and 20 °C [77]. The minimum moisture threshold has been introduced as the metrics having the most significant impact on predicting fungal growth [11, 78]. It represents the MC below which fungal decay cannot be initiated, and is typically given at an mass loss 2%.

Available hydroxyl groups are the predominant sorption sites for water molecules in wood. Their presence in hemicellulose is twice as high as in lignin and four times higher than that of cellulose [79]. It has been reported that cellulose can endure structural changes while wood is desiccated [80, 81]. In the literature, it was hypothesised that lower desiccation rates would give enough time for amorphous cellulose to crystallise, rendering the substrate less hydrophilic. On the contrary, higher rates of desiccation may lead to more prominent amorphous cellulose and more hydroxyl availability. Lignin, hemicellulose and pectin interrupt cellulose crystallisation, which reflects on the higher crystalline content towards the S3 cell wall layer [82]. Because hemicellulose has a softening point at room temperature and 18–20% MC [74], while lignin softens above 60 °C at a minimum of 15% MC [83], and cellulose is hardly softened in its crystalline conzuration (200 °C), higher thermal treatments may result in hemicellulose degradation, thus rendering the substrate less hydrophilic [84].

Finally, the amount and quality of water in the SSF system varies during fermentation. Fungal oxidative decay of polysaccharides frees water along with CO_2:

$$(C_6H_{10}O_5)_n + 6n(O_2) = 6n(CO_2) + 5n(H_2O) \tag{20.1}$$

where $C_6H_{10}O_5$ is the repeating unit of glucose-based polymers, such as found in starch or cellulose. As bound water is also released during decay, and while substrate dry mass loss occurs, the MC increases if the system is closed. The activity and location of water in SSF systems are are then necessary information to predict fungal development. They are of primary interest to design and produce MBC. To the best of our knowledge, the role of water in MBC production has not yet been reported. The optimal moisture content for laccase, LiP and MnP activation is consistently reported to be 60–75% [85–88].

20.5 Regulating Enzymatic Activities

MBC protocols can be adjusted based on the chemical profile of locally available wood resources and selected fungal species to achieve various design goals. This an be approached by pH and temperature regulation, and monosaccharide, nitrogen and metal ion supplementation. Optimal ligninase activity was reported at pH 4.0–5.0 and 30–35 °C [85, 86]. The pH regulation can be achieved by the addition of $CaCO_3$ in the substrate as a buffering agent to avoid acidification from CO_2 production. Gypsum supplementation has been reported in MBC cultivation [89]. While its use has no impact on the substrate pH, it contributes to adding calcium to the medium. This is useful for pectate lyase activity if the wood has a low Ca^{2+} content, which acts on the 1–4% share of pectin in wood. Varying the C:N ratio has also been shown to lead to an optimum of 20:1 [85]. The form of the C- and N-rich additives has been reported to have an impact on specific enzyme activation. Maltose and urea addition was reported to increase LiP and MnP production in a *T. versicolor* species, while glucose and yeast resulted in increased laccase production [87]. A similar investigation with a *G. lucidum* reported that the use of beef extract increased LiP production by approximately 30% as compared to using yeast extract, peptone or urea, and sucrose had a significant effect on ligninase activity as per other sugar types [85]. Wheat bran is a frequently used substrate additive in SSF. It contains high contents of β-glucans, xylan, cellulose, lignin, B vitamins and mostly minerals (magnesium, phosphorus, manganese, zinc and iron) [90]. The success of this supplement [91] may be related to its mineral contents, especially manganese as it is a limiting compound for MnP and VP activity [92]. Supplementation of substrates by $MnSO_4$ has therefore been shown to result in an increase in ligninase activity in *S. commune* [86] – most probably by synergistic action of enzymes. Biofuel production aims at efficient extraction of sugars from wood. Selective delignification capable fungi have been extensively investigated in this context. The addition of phenols has been reported to be considerably promoting ligninase activity [93]. Syringic acid has been shown to increase LiP and laccase activity, and gallic acid for MnP in the same study. With an optimised substrate, the authors reported increase of 2.4 to 45.5-fold across the diverse ligninase activities, leading to a 2.3-flod improvement in lignin decay and 7.1 in selectivity value (selectivity is expressed as the ratio of lignin degradation over cellulose and hemicellulose degradation) [94]. The idea of highly selectively degrading lignocellulosic substrates is a very promising track for MBC research. The addition of H_2O_2 to the substrate, necessary for ligninase activity, has also been investigated with significant results [95, 96]. $CuSO_4$ supplements have been shown to increase laccase activity [93]. The use of surfactants such as Tween-80 has been reported to increase ligninase permeation through the cell wall [97], and enhance the solubilisation of lignin and improve hydrolytic activities [98], and even proved useful more recently in promoting all ligninase activity, at a 4 wt% optimum for *G. lucidum* fermentation [88]. With an approximate 80% content of starch, 12% proteins, 3% of other monosaccharides (arabinose, xylose, galactose, glucose) and traces of ash (minerals) [99], white and whole wheat flour constitute useful additives to promote cellulase and hemicellulase activities. Moreover, the use of spent coffee ground is a widespread decay promoter in MBC craft. It is composed of 39% hemicellulose (predominantly mannose and galactose), 24% lignin, 17% proteins, 2% fat and 1.3% ashes. Although it has a high lignin content, coffee grounds contribute to increasing nitrogen and mineral content in the substrate, additionally to glucose polymers. Coffee silverskin, a by-product of the roasting industry, has a quite different chemical profile: 24% cellulose, 29% lignin, 19% proteins, 17% hemicellulose (primarily xylose) and a remarkable 5% ashes [100]. Phenolic compounds present in both substrates have been investigated for their antifungal abilities against white-rot fungi. Using a *T. versicolor* species, it was reported that the supplementation of 3% coffee silverskin extract resulted in about 65% growth inhibition, while 1% caffeine led to a complete inhibition and 1% spent coffee ground in about 20% inhibition [101]. This is related to the introduction of phenolic antioxidants such as caffeine, caffeoylquinic acid lactone and feruoylquinic acid [102–104]. Furthermore, laccase treatment of lignocellulosic substrates has been shown to increase lignin antioxidant activity [105]. Finally, the addition of phosphorus and calcium in the form of superphosphate ($CaH_6O_8P^{2+}$) has proved to increase decay yield. Varying the supplementation from 0.025 wt% to a 0.3 wt% optimum in wheat straw, lignin degradation increased by a 4.5-fold, while cellulose, hemicellulose and protein degradation increased by a factor of about 2.4, while enduring *P. ostreatus* decay [106]. A higher optimum of 0.5 wt% superphosphate addition has been reported for alkaliphilic white-rot *Coprinus* spp. [107], and even 1.0 wt% with *T. versicolor* [108].

20.6 Mycelium Mechanical Properties

Mycelium acts as a binder between wood particles. It does so by degrading lignocellulosic compounds and producing a dense hyphal network that interlocks dispersed particles. The binding between two particles is not distinctly strong, but the very high redundancy in the composite can result in a mechanically valuable material. It has been reported that a *P. ostreatus* mycelium would be more than two times stiffer than a *G. lucidum* one when both cultivated on a cellulose medium [109]. Authors comment on this difference with regard to the higher protein and lipid content in *G. lucidum* that can act as a plasticiser. Furthermore, in this study a set of experiments grown on a PDB-cellulose medium has been compared to the previously mentioned results, and it was found to decrease the Young's modulus of the *G. lucidum* mycelium film by a factor of 3 while increasing its elongation at break by 2.3-fold. This corroborates their previous comment on plasticisers, and suggests that nutrition medium tuning can help composite behaviour. This study suggests that the matrix phase of the composite can be made closer to an elastomer or stiffened by varying protein, lipid or alcohol contents. Similar findings have been reported [111]. Isolated mycelium mechanical behaviour has been investigated in order to model it [112]. This referenced study relies on the characterisation in tension and compression of a pure mycelium. At the resolution of hyphae, researchers have identified three main stages of response of a ligninolytic fungal mycelium: linear elastic behaviour at small strains (hyphae bending); fibre buckling and local structural collapse or densification at larger strains; rapid stiffening associated with full compaction and large number of inter-fibre contacts. To fit their finite-element analysis simulation model, a hyperelastic model was used in this study as mycelium undergoes large displacements in its elastic behaviour and results in a non-linear stress-strain response. Young's moduli are situated in the 0.6–2.0 MPa range for a density in 30–50 kg/m^3, the yield stress in 40–80 kPa and ultimate tensile strength in 100–300 kPa [110]. The results are presented in Table 20.4.

Table 20.4 Tensile behaviour of mycelia from *P. ostreatus* (PO) [109], *G. lucidum* (GL) [109] and an undisclosed species (ND) [110]

Tensile behaviour	PO[a]	PO[b]	GL[a]	GL[b]	ND[c]
Young's modulus (MPa)	28	17	12	4	0.6–2.0
Ultimate strength (MPa)	0.7	1.1	1.1	0.8	0.1–0.3
Elongation at break (%)	4	9	14	33	–

[a] Cellulose medium.
[b] Cellulose and PDB medium.
[c] Undisclosed medium.

20.7 Substrate Mechanical Properties

Most commercially valuable woods have a Young's modulus of 5.5–15.7 GPa at 12% MC, and 4.4–12.3 GPa when freshly cut (green) [113]. The distribution of these performances is similar in gymnospermous and angiospermous woods. Wood particles are therefore important load-carrying members of the composite system as compared to mycelia elasticity discussed previously, and reduce the magnitude of stress experienced by the mycelial matrix. The angular shapes of wood debris contribute to crack initiation and the dewetting behaviour of the larger particles is a principal contributor to damage nucleation in such a two-phase particulate composite, although they contribute to composite stiffening by matrix shielding. Furthermore, the shape, nature and distribution of particles in two-phase particulate composites has been shown to have a substantial influence over the load transfer between members, and hence their overall stiffness [114]. Moreover, while lignin is a primary contributor of strength parallel to grain, hemicellulose supports compression strength perpendicular to grain. Its decay greatly affects the structural integrity of wood and its hardness [115].

In particle studies of granular materials, shape parameters such as flakiness/flatness (ratio of particle thickness to width), elongation (length-to-width ratio), sphericity (deviation from a sphere geometry) and roundness/angularity (measure of angular sharpness) have been investigated [116]. Without matrix phase (the stiffest reported pure mycelium of a *P. ostreatus* strain in 20.6 had a Young's modulus of 28 MPa, 178–534 times more elastic than woods), studies have reported that a 3:1 ratio of flaky to bulky particle content would be an approximate optimal for shear strength (depending on the system of study). This was related to cohesion increase under stress due to particle interlocking. Furthermore, it was reported that a higher particle angularity leads to a decrease in elastic modulus, while ultimate strength increases. The shear strength was reported to follow the same dynamic, increasing with particle angularity. Increasing flakiness and angularity leads to increased cohesion and subjection to higher abrasion. This leads to damage accumulation under repeated loads, thus leading to strain accumulation [116].

While wood particles are of common use in MBC, other more elastic substrates have been used such as nonwoven cotton fibres [117]. A few studies have investigated the addition of non-organic aggregates to a lignocellulosic substrate to improve its stiffness, such as with carbonate sand [118] (which also acts as a pH buffer being mainly composed of $CaCO_3$) and sand and gravel [9]. It was reported that supplementation of a cotton and wheat bran medium with 37.5 wt% carbonate sand resulted in a 1.6-fold stiffening, and a factor 4 increase in ultimate compressive strength, while density increased by 27%.

20.8 Composite Mechanical Properties

The large number of influential variables on MBC mechanical properties leads to the emergence of a variety of methods for tuning them. While common practice leads to an elastic modulus of an approximate maximum of 2 MPa, post-cultivation heat-pressing has been reported to lead to moduli in the range of 35–97 MPa, and flexural moduli in the range of 34–80 MPa [117]. The use of particle-additive strategies, additionally to a wheat bran supplemented medium made of cotton stalk, has proven to have a significant effect: an elastic modulus of 48.5 MPa was reported with 37.5% carbonate sand addition with *P. ostreatus*, while a non-supplemented control in this study reached 30.3 MPa [118]. A second study reported an elastic modulus situated in the 39–60 MPa range for a density in 240–265 kg/m^3 [7]. Authors of the latter are elusive about production details, but nonetheless indicate the use of Alaska birch shavings (*Betula neoalaskana*), millet grain, wheat bran, calcium sulphate and the addition of a natural fibre. The specifics of the performative batch are not disclosed, but a pre-colonisation was performed prior to moulding. The performance increase does not come from a single factor considering the experimental plan. The considerable use of fibres (of undisclosed quality), grains, brans and $CaSO_4$ leads us to understand this as a hybrid improvement using both substrate-based reinforcement with the fibres and additives. Other strategies have been investigated, such as with the introduction of SBR Latex, a bonding agent for the construction industry. With a 5% addition of SBR Latex and 0.5% silane coupling agent to a cotton seed hull medium, the cultivation of a *P. ostreatus* species displayed a 2.39 factor increase in ultimate strength with a 27.6% density increase [119]. The resulting environmental impact has not been discussed in the study. The addition of another material with high environmental impact [120], cellulose nanofibrils (CNF), has been reported [121]. Between two specimen groups, the optimal supplementation of CNF was reportedly 2.5–5 wt%, with a contribution in Young's modulus of a factor 2.56–5.6 and 2.8–7 in ultimate strength. In this study, the effect of densification on composites embedding a 2.5 wt% CNF part was reported: densifying from 300 to 600 kg/m^3 contributed to increase the yield strength exponentially to a factor 17, while the stiffness increased 14.38-fold.

An extensive study of the properties in tension of densified MBC reported that elastic moduli situated in a range of 3.0–13.0 MPa; these values resulting in 24 days cultivated *T. versicolor* mycelia on wheat bran supplemented beech sawdust [117]. For this specimen group, a flexural modulus of 9.0 MPa was reported. The state-of-the-art in MBC research largely considers monolithic and homogeneous composites, besides a few study groups investigating jute type materials in sandwich composite reinforcement and composition with wood panels [45, 122–124].

Across the state-of-the-art, we can identify three main research strategies: *densification* (by composite packing, cold- or hot-pressing), *composition* (by introducing structuring elements such as fibres and aggregates) and *supplementation* (by chemical tuning of the substrate). These are substrate-focused strategies, targeting mycelium properties tuning in the case of supplementation, while the very choice of the fungal species can also result in radically different cultivation lead time and mechanical properties. Genetic modification of fungi could promote specific enzymatic activity or modify the composition of its cell wall to help improve performance. However, the investment cost and ethics of this practice may lead to prioritising more energy-efficient and affordable techniques.

20.9 Analysis of the State-of-the-Art

So as to analyse the effect of strategies in the MBC state-of-the-art and to present a comprehensive mechanical review for it, we collected reports of elastic moduli and ultimate strength in compression, tension and flexion, along with principal substrate information, species and density [7, 45, 117, 118, 121, 122, 124–135]. There are 105 data points gathered from 18 journal and conference articles. These include articles reporting on all or some of these metrics: strength and/or stiffness in compression, tension and/or flexion; 19 data points had no density reported. In the following data visualisations (Figs. 20.5, 20.6, 20.7, 20.8, and 20.9), only the reports with sufficient data are rendered.

The distribution of species to substrates reports is heterogeneous. For instance, we can observe in Fig. 20.5 that no peer-reviewed report has been made of hemp cultivated *G. lucidum* as per mechanical behaviour. This figure does not consider all substrates and supplements included in MBC, only the primary one.

We present the specific moduli and strengths as per substrates in Fig. 20.6, and specific moduli and strengths as per species in Fig. 20.7. Larger marker size indicates

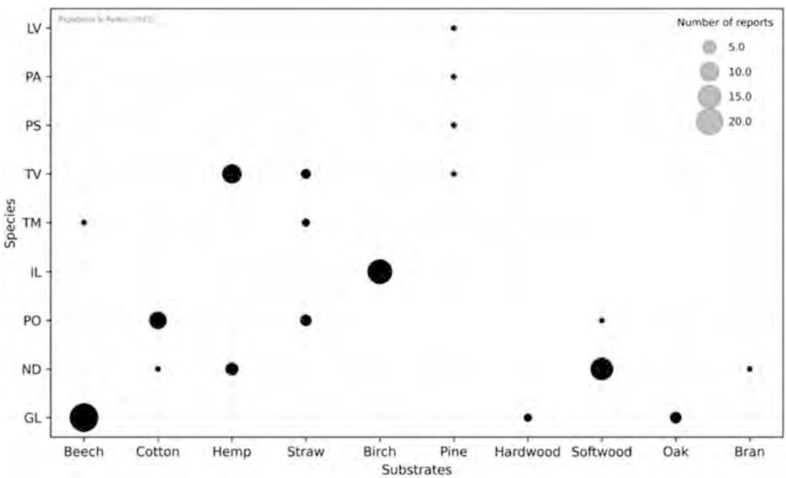


Fig. 20.5 Reports of principal substrates and species. GL: *Ganoderma lucidum*, ND: undisclosed, PO: *Pleurotus ostreatus*, IL: *Irpex lacteus*, TM: *Trametes multicolor*, TV: *Trametes versicolor*, PS: *Pycnoporus sanguineus*, PA: *Pleurotus albidus*, LV: *Lentinus velutinus*

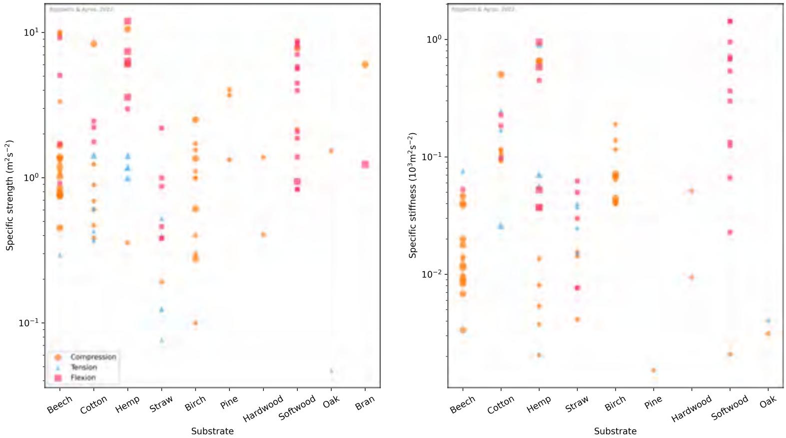


Fig. 20.6 Specific strength and stiffness per substrate. Larger markers correspond to composition strategies

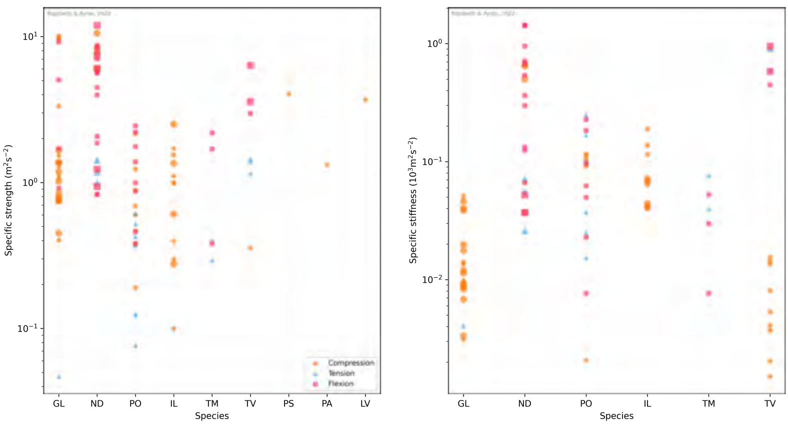


Fig. 20.7 Specific strength and stiffness per species. Larger markers correspond to composition strategies

a composition strategy. We classified MBC having structuring fibres, textiles, inorganic particles, CNF and long fibre balls. Only a few reports on MBC tension modulus (14 data points) and strength (15 data points) have been published. From biochemical review presented above, we can be surprised by the mechanical

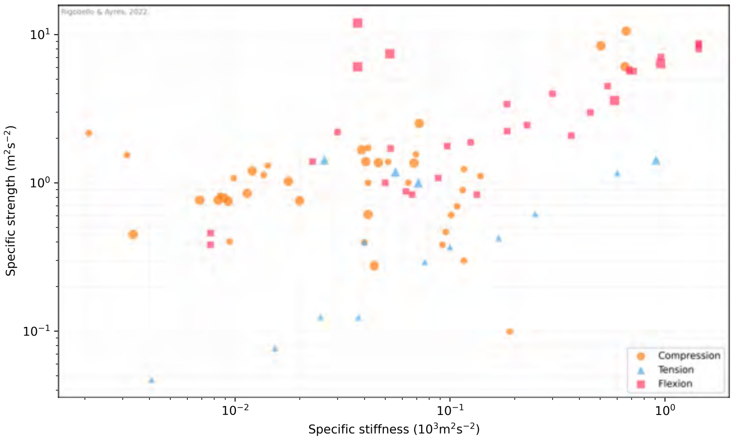


Fig. 20.8 Specific strength as a function of specific stiffness. Larger markers correspond to composition strategies

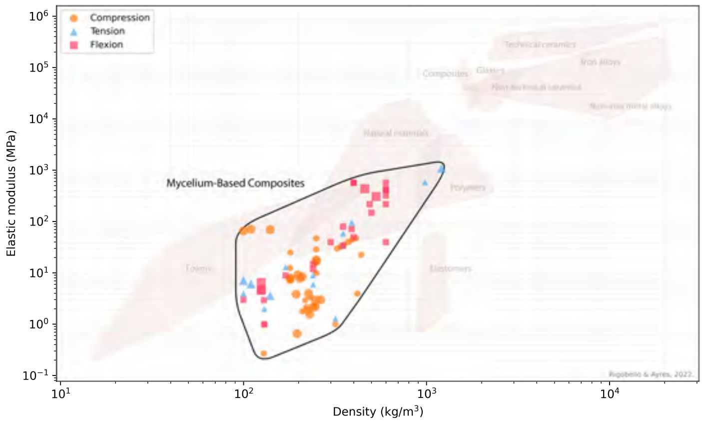


Fig. 20.9 Elastic modulus as a function of density. Larger markers correspond to composition strategies

competitiveness of softwood in Fig. 20.6. It should be noted that these specific reports are of CNF-embedding MBC, thus supplementing softwood with cellulose that is accessible to the fungus without a lignin barrier, having been pressed at an undisclosed pressure at 180 °C for 15 min [121]. Densification of lignocellulosic particles by hot pressing may result in an increased mechanical interlocking of the particles, along with chemical binding from solubilised compounds [83]. Furthermore, in Fig. 20.7, we can identify the undisclosed species in use to be the most performative with regards to specific stiffness and specific strength. In Fig. 20.8, we present the specific strength as a function of specific stiffness in compression, tension and flexion. We can notice in this figure a departure from the state-of-the-art of a specific study in compression [124]. The specimens of the latter were embedding a surface-binding fabric and packed cotton-ginning waste and hemp pith of undisclosed qualities. The reported specimen densities are of 100–140 kg/m^3. Without further details on the cultivation method, and with the tests having been conducted with only 3–4 replicates, we can hardly conclude on the causes having led to these outstanding values. With critical methodological details being undisclosed, this study is irreproducible and, as such, serves solely as advertisement material.

We present an Ashby map of state-of-the-art MBC data for elastic modulus as a function of density in Fig. 20.9. We can notice that MBC represent a large functional area, mostly situating in between foam and elastomer materials, with reports extending the group span to performances typical of natural materials – the upper-right cluster of flexural modulus reports values typically stiffer than cork and leather. Punctual reports of tensile modulus in the upper-right MBC area would be of a similar stiffness than polyethylene (PE) or PE polymers. The span of the MBC area reflects the three development strategies: densification leads to clear tendency towards increasing the stiffness and strength of MBC for construction materials; supplementation is less distinct at the moment in the reports, but the three data points in the upper-left MBC area are representative of its effect as shown in Fig. 20.8; composition strategies are distributed across the MBC area and reflect a wide variety of inserts and resulting mechanical behaviour. Composition has been demonstrated to lead to significant effect on MBC behaviour in failure control or stiffening for instance [45].

20.10 Conclusions

This review focuses on three aspects: fungal identity (fungal species enzyme array), nutrients (substrate chemical profile and wetting characteristics) and substrate architecture (substrate aggregate characteristics, density and composition). From the enzyme review, we have come to understand that the substrate fitness and mechanical behaviour of mycelia can be adjusted by supplementation. This can be achieved by pH and temperature regulation, and monosaccharide, nitrogen, metal ion and $CaCO_3$ supplements could be systematically investigated in the future as per initial substrate pH and its variation during fermentation. Lignocellulosic substrate selection can also have a significant effect, with non-supplemented softwoods being chemically unfit for optimal white-rot decay. The use of sycamore or ash wood could be investigated in the future as they display higher syringyl monomer contents. The C:N content of sycamore being 401, supplementation may be necessary for optimal fermentation. Furthermore, incubation temperature is commonly fixed throughout the fermentation process below 30 °C, which promotes specific enzymatic activities (LiP, MnP, DyP, xylanase, pectate lyase, GH1 β-glucosidase and

GH3 β-glucosidase). While investigating exothermic heat generation and dynamic temperature control related to species, genome distribution may lead to both a widening and deepening of the available material qualities and versatility.

The effect of water activity upon MBC colonisation-related performances, such as mechanical response, has not yet been reported in the state-of-the-art. Considering reports in SSF studies of several fold increase in colonisation rates, investigating the effect of water activity in MBC may result in longer production times and performance. Moreover, qualifying the effect of hydroxyl group availability in hemicellulose and cellulose, depending on wood processing history, could lead to further medium optimisation. We expect this particular aspect to be specifically useful for paste-like substrate design, such as that found in mycelium-related 3D printing.

Of course, stimulating the focus of future research towards such specific aspects may result in a higher entry barrier for a technique that has a strong interest for its affordability. The use of Fourier transform IR or other spectrometric process by experts within a network of amateur practitioners could be useful to adapt cultivation strategies constructively to local contexts; the qualification of an available substrate may lead to specifying necessary processing, supplementation, species selection and/or relevant composition strategies. While industrial and academic research may result in defining a panel of protocols for the selection of prospective materials, situated citizen craft practices based on locally available materials could coexist.

Across the MBC state-of-the-art, we have identified three principal substrate-oriented design strategies: densification, supplementation and composition. The latter covers a variety of strategies, from CNF introduction to uniaxial reinforcement. These will need to be further investigated to allow an in-depth understanding and serving a goal-oriented material development. It is important to note that most experimental methods are heterogeneous, may not be statistically significant or only partially disclosed. A structured approach to developing material designs along with assessing them will most likely result in the concentration of the state-of-the-art around functional poles in the future, and will help to drive material development in depth. Additionally, future reproducibility studies and critical methodological reviews of MBC studies may both improve the overall market readiness and public adoption of MBC solutions. This can further valorise the wealth of material, design and semiotic properties that the versatility and affordability of MBC systems support.

In this chapter, we have identified the principles driving MBC design and engineering, and have attempted to make these comprehensible. Beyond this focus on the design and production, MBC engage broader discourses related to resource sourcing, circular economy and sustainability. Because MBC have a higher vernacular potential [136], they can be made of recycled substrate, or materials locally and seasonally available. MBC also benefit from unique aesthetics as a result of the phylogenic expression of fungi through craft and substrate design [137]. We hope that the on-going uncovering of the principles for predicting MBC performance contributes to an engineering practice that may satisfy industrial needs (which detrimentally often actualises in patents [138]), but also fosters a rich, open, sustainable and creative citizen craft.

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F Conference articles

F.1 Fragile computation: rethinking information technologies to foster situated ecologies.

Fragile computation: rethinking information technologies to foster situated ecologies.

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Keywords.

Milieu, Design theory, Ecology, Natural resources, Terroir

Abstract

From the 1950s on, the informational paradigm revived ecological holism and supported a vision of total inference. The practical implementation of the novel communication technologies at the time made possible the globalisation of the economy. The latter was a cornerstone of unprecedented economic growth and welfare, but by globalising the resources that technical objects and systems relied upon, it seeded dramatic more-than-human inequalities that accelerated global biodiversity depletion. While modern technologies evangelists promoted the idea that we could assess this phenomenon thanks to complex modelling and advanced engineering, the implementation in a global economy introduced a distance in the structuration of technical artefacts' mode of existence, that is by geographically generalising the natural resources at stake. Furthermore, the latter advent of the web in this economic paradigm augmented the erosion rhythm of the geographic situatedness and structurally nurtured technological placelessness. Instrumental to structurally implement sustainability in design activities, we analyse here the notion of geographical situatedness in technical objects. By looking into case studies of terroir and vernacular architecture, this work suggests that the prism of locality, in that it makes situated natural resources a non-negotiable trait for the mode of existence of technical objects, bears a high potential for increasing communities' resilience by resources economy. Throughout terroir definition and analysis, we identify craftspersonship as a keystone to further reduce the gap between users of technical objects and underlying resources mobilization, thus contributing to environmental care and awareness. Finally, we illustrate the use of terroir qualities with an alternative Information technology using living and dried fungi. By resorting to highly affordable fungal materials and crafts and developing a design that specifies on-site at the architectural scale, we bridge the use of novel materials and knowledge with a structural ecological affordability.

1. Introduction

Built upon the innovations in communication technologies from the Second Industrial Revolution and their widespread post-war democratisation, cybernetic thinking and its focus on system regulation offered a relay to a debate that had been happening in Ecology since the early twentieth century regarding the validity of ecological holism and individualism (Simberloff, 1980). Just over 200 years after the beginning of the First Industrial Revolution, and with a rising global concern about biodiversity depletion caused by human activities, ecological holism was re-actualised by the Gaia theory (Lovelock, 1972). This school of thought notably supported the idea that natural resources can be described, controlled, and exploited rationally and scientifically in the cybernetic paradigm. But natural resources management has always been central to the establishment and maintaining of power relations, subjected to mediation between various human societies interests. What a power structure regards as a legitimate resource is guided by its ethics and values, but leads to an objectification of the resource and the need for it to be managed by rational methods in the modern era. The acting out of this power relationship can be seen from the birth of agriculture and human settlements, with mechanisation and industrialisation from 1760 on, arguably initiating the global imbalance at a scale that is currently referred to as the Anthropocene (Charbonnier, 2020).

In the present informational paradigm, it is argued that recursive environmental modelling, at the fine resolution of the data we can acquire, would theoretically allow humans to build trustworthy discrete models of complex environmental phenomena. Hence, we could increase our knowledge, model fitness, and event predictions from studying complex systems, and plan, scientifically, to support decision-making. This is an epistemological realism (Kitchin, 2015); all the data that is being captured of the world forms our knowledge of it. But through its actualisation in the modern socio-technical paradigm, this epistemological framework tends to relegate the non-humans actants composing the land (which provides resources for the becoming of technical systems) to a status of simplified resource - objectified through a process of approximated digital modelling (Winsberg, 1999), and subjected to the enactment of human-centred power structures. Catastrophic industrial events and biodiversity depletion acceleration in the modern era particularly illustrate an effect of this objectification, which is the distancing of human actors - users, inhabitants - from the resource extraction that allows the existence of the technical object at hand.

With a particular focus on information technologies, this article proposes an analysis of the structuration of the association chains of technical objects to their resources' geographies through the notion of technical milieu (Leroi-Gourhan, 1945) and supported by an understanding of placelessness. We find that placelessness can be implemented through the process of translating human actors' intent to non-human objects (Latour, 1990) by elongating association chains and black-boxing, thus supporting a delegation of the associated natural resources management. After analysing terroir and vernacular architecture case studies as technical milieus that are deeply interwoven with - and even derived from - their natural milieus, we proceed to exemplify the recourse to the structural qualities of these by applying them to an alternative Information Technology - a living and architectural fungal automaton.

2. Placelessness

The principal aim of this study is to offer an analysis of the way geography, and its related natural resources, are associated with technical objects. Before proceeding to this analysis, we need to understand what placelessness encompasses. French economist Pierre-Noël Giraud offers a useful perspective; he suggests the economy is composed of two principal jobs poles: nomad and sedentary (Giraud, 2015). Nomad jobs are characterised by their independence from the land; they represent a set of competencies with which an individual can virtually relocate and do a similar job. These jobs (persons, competencies) are globally competitive, across nations. An IT engineer from India could have the same competencies to a German one; their jobs are in competition and the land is irrelevant. Such job profiles are instrumental to the globalised model of production and consumption; they are typically represented by persons with a higher education (such as master level for instance) and represent a large economic volume in exchanges. The higher buying power that nomad jobs benefit from is spent locally, on the land they inhabit towards sedentary jobs. This class is the inverse of the former in that the competencies they represent are always dependent on the territory they inhabit. A non-English speaking baker for instance, or the whole agricultural economy, are good representatives. This model is obviously dynamic and subject to the availability of education programs for instance, but it provides valuable readings. In the period 1990 – 2010, India as a developing country had a slightly increasing ratio of nomadic to sedentary jobs between 10% and 15% of the active population. Over the same period, the ratio of economic added value per nomad job to the added value of sedentary job went from 220% to 170% (Giraud, 2015). The latter metric is indicative of a reducing, yet high level of national inequality between these two poles. The effects of this inequality are illustrated by the long-standing and on-going conflict between global corporations (such as chemical, food and beverages industries), and Indian farmers. The resorting to High-Yield Variety seeds and monoculture in India, supported by global corporations and presented as a fix for farmers to supply the local demand, have caused dramatic environmental damages and weakened the resilience of these farmers (Bhatia, 2020; Dutta, 2012). The world-wide investment on nomadic activities to serve global competition has been a contributor to many geographic areas suffering from biodiversity depletion and long-term loss of equilibrium in agricultural health of biological milieux (Akram-Lodhi *et al*., 2009). This has reached such a level that society felt it deserved a specific geologic era name: the Anthropocene (Crutzen, 2006).

A major and increasing part of the competitiveness of nomad profiles relies on access to the web. The networks of knowledge, communities, or even cultures that the web allows access to, find indubitable benefits in terms of autonomy and empowerment of communities. This position underpins major efforts to bring innovative web infrastructure to remote communities, especially via low-orbiting satellites (Massey *et al*., 2020). While an increasing number of web access opportunities are being granted to more sedentary communities, the recourse to the web itself erodes further the relevance of the land. In fact, the supposed placelessness quality of the web (from a user perspective) has supported a recent and tremendous shift in geopolitical power structures (Srnicek, 2017). Companies such as Google, Apple, Facebook, Amazon, and Microsoft (GAFAM) have developed a power relationship that is beyond that of some nations, as they occupy a strategic position in the access to web resources. This shift in power is of particular significance as it was strong enough to disrupt the annuity circumstances of territorial actors (Beaude, 2012), such as traditional taxi services. In the actualisation of a techno-utopian vision for the web, the service economy got boosted by web platforms (delivery services, on-demand transports, etc.). These services are using the convenience of web placelessness and provide high-end offers to their users. By typically automating administrative tasks of information communication, platforms make the delegation of a “pain point” feasible to the user with a service fee (Kenney & Zysman, 2016). On top of the intrinsic web placelessness, delegation is built-in recursively and further contributes to eroding the relevance of the land. In this framework, the association of technical objects and systems aggregates, expands and autonomises from the user (as the provided service is by definition a delegation of expertise), and geography. In the perspective of full coverage and universal access to the web, should we desire to mitigate and anticipate anthropocentric environmental depletion and smarten up our natural resources' management, we suggest that assessing the intrinsic placelessness to become can be done by inducing a cross-pollination of nomad - sedentary activities. This means understanding and implementing the situated structure of geographic resources into the mode of existence of technical objects and systems, to provide a framework for sustainable technological development.

3. Of fragile milieus

a. Techno-geographic milieu, inverted

The role of geography in relation to the mode of existence of technical objects, has been investigated previously; building on the notion of *technical milieu* – a mediation between the corporeal and/or societal internal milieu and the external milieu – Simondon introduced the notion of *techno-geographic milieu* (Simondon, 2016). If a technical milieu mediates between subject and phenomena, the techno-geographic one is conceptually made dependent on its geographic context, strengthening the link between the technical object and its external milieu. The latter is further articulated such that the technical milieu exists only if and when the geography allows it. Simondon refers to this interdependent association of milieus when a multitude of functions are being integrated in such a way. He uses the Guimbal hydroelectric turbine as an example, which generates an electric current from converting hydraulic pressure whilst using the water flow as a coolant. Expanding upon this example, the turbine can be adapted to virtually any system looking to convert hydraulic pressure to an electric current, with the specificity of having an extended compactness (a reason that motivated this design), and scope of usage as it embarks a regulating cooling function. The turbine therefore functions thanks to its integration into an external milieu, but the external milieu is merely a well understood reservoir of functionalisable resource to the artefact. Although it is evident that the milieu is associated with the object in more than one way, we argue that the inverse is not true. In *On the Mode of Existence of Technical Objects*, Simondon investigates the genetic evolution of technical objects in the section where he describes the techno-geographic milieu; through the concept of *hypertelia*, of design trait of exaggeration and supposed inefficiency, Simondon sets himself on a path to investigate the modern conditions of mutation and selection of design genes and praises the Guimbal turbine for what it serves to this research: modern efficiency. If the techno-geographic, or association quality of the Guimbal turbine is understood as: *the technical milieu utilises in more than one way its external milieu*, then the proposition that we seek to increase the weight of the natural milieu in the existence of the technical, is the inverse: *the technical milieu is dependant in more than one way of its external milieu*. While the techno-geographic milieu rewards genius in anthropocentric efficiency, the inverse proposition – from *utilising* to *depending* – implies that the technology cannot exist outside of its design territory. What appears as modern nonsense finds a tremendous echo in premodern and non-modern designs.

The modern era is characterised by an increase in technological availability in all domains. Direct and visible effects include successive demographic transitions from the nineteenth century on, marking the effective conversion of a precarious society – in terms of life expectancy and infantile mortality – to a stable one. The conditions of validity of the modern paradigm have been extensively discussed (Charbonnier, 2020; Latour, 1993). With the technical progress in the areas of communication and transport throughout the industrial age, the globalisation of resources was made affordable, and the communication reach extended almost beyond all geographical limits. The resource availability to those in power is guaranteed, famously making annual human consumption well over the limits of annual environmental regeneration in the past years[1]. The apparent global abundance of resources supplies a pattern of consumption that is disarticulated from the local availability of them. While not having the structural access to global resources, premodern and non-modern human societies relied on finite and potentially regenerating supplies, that is in terms of natural, technological, and human resources. More often out of need rather than desire, at least for a time, non-modern technical milieus that have been developed as such have a specific array of qualities in relation to the territory. We are illustrating our research with two examples: terroir, and traditional vernacular architecture.

b. Dependencies in terroir

In 2005, INRA, INAO and UNESCO[2] agreed on a definition of terroir as the following: "A terroir is a delimited geographic space defined by a human community that builds throughout its history an ensemble of distinctive cultural traits, of knowledge and practices, based on an interaction system between the natural milieu and human factors. The know-how at play reveals an originality, confers a typicity and allows a recognition for products and services that originate from this space and for humans that inhabit it. Terroirs are living and innovative spaces that cannot only be assimilated to tradition." (Prévost *et al.*, 2014). Although the term originates from agricultural practices, where a product quality encompasses a complexity in taste reflecting its peculiar and specific territory revealed through advanced craft, it is remarkable that this definition opens the scope of the qualificative beyond edibles. In terroir, the environment is so associated and intertwined in the mode of existence of the object that the methods and materials are protected from modern normative food safety demands – with conditions of control of the

[1] www.footprintnetwork.org. Accessed April 9th, 2020.

[2] INRA: Institut national de la recherche agronomique; INAO: Institut national de l'origine et de la qualité; UNESCO: United nations educational, scientific, and cultural organisation.

finished product. The fauna, flora and microbial communities are a core part of the milieu with instances such as usage of woods in aging, or recourse to naturally occurring ferments. While the existence of the products and services is mostly inherited out of necessity, the proposition that the *technical milieu is dependent in more than one way of its external milieu* applies nevertheless in protected bills of specification for the product.

Far beyond a romantic pastoral notion, terroir has been a key axis of economic development for the United-Nations in the South from the mid-1980s onward after a general movement away from projects emphasising technical aspects of rural development in Sahelian West Africa. At the time when there was a rising recognition that the impact of agricultural intensification over natural resources and the environment had failed to be considered, Gestion de Terroir[3] projects started to be implemented (Toulmin, 1994) with three main approaches (Dalal-Clayton *et al*., 2000):

- Natural Resources Management: focusing on the physical improvement of the natural resource base through existing institutions,
- Institution building: creating and supporting community-based institutions,
- Local development: organising communities and the transfer of financial responsibility for rural land use planning to the committees.

Although its implementations have encountered critiques or shortcomings (World Bank, 1998), we can appreciate that the Gestion de Terroir approach acknowledges a strong focus on community-led local institutions and know-hows for fostering both the autonomy regarding natural resources management and promoting the resulting craft and cultural identity.

The notion of craftspersonship is central not only to the terroir, but also to vernacular architecture. Vernacularity refers to the affordability of resources to a community, and it helps to define the locality from technological and knowledge availability – whether it has access to mechanised vehicles influences the resources reach, for instance. Vernacular architecture traditionally relies on a distributable potential of the expertise and craft among the community to improve its resilience. Shared constructive methodologies that have been designed and tested by the community allow individuals to be able to both design their works with confidence in their safety, but also to perpetuate the reproduction of design traits indicative of social belonging. Used materials are traditionally subject to rather rapid erosion depending on the location and traditions; the shared craft know-hows and historical experience in maintenance can form opportunities for ritualised caretaking of the works, bridging the gap of relative fragility with community perpetuation. Furthermore, the definition of terroir that we cited earlier does comprise such practices: the geographic space is defined by a community production based on an interaction system with the natural milieu and human factors. Throughout these two contexts, we understand that with terroir - the inverse to the Simondonian techno-geographic milieu - the resilience of the community is as great as its technological fragility[4]. Conversely, the Shintoist tradition of Shikinen Sengū-sai consisting in rebuilding temples every twenty years according to original construction methods and materials illustrates another case of embedded fragility. The temples are known to be rather ornamental and sturdy; the process of reconstruction is a celebration of continuity through reproduction rather than conservation (Coulmas, 1994). Here the introduction of a structural fragility via the ritual both supports a specific religious ontology, but as for what concerns us, community fostering too. In contrast to traditional vernacular architecture, it is not the limits in material accessibility or material wearing that motivate the introduction of a fragility.

Fragility can be a parameter to balance the design of a technical milieu and its dependency to the internal and external milieus. Introducing fragility into a technical system is a way to shift the power from the authors of black-boxed knowledge that is, or can be, embedded into a technical object, towards collective intelligence. This intelligence is in crafts and their knowledge transmission methods. This is illustrated in traditional vernacular architecture with oral and written traditions, but also in agricultural terroir as discussed earlier. The cost of making rammed-earth constructions or supporting the structuration of a terroir in its communal sense (that is by investing in craft education for instance), although it might make the central object fragile – subject to eroding and wearing – confers an autonomy to the local community in managing the natural resources. This autonomy, as opposed to delegation via the multiplication of interfacing technical milieus, contributes to form a terroir by reducing the distance from the user to the resources and by making the latter a non-negotiable trait of the mode of existence of the central technical object or system. Paradoxically, the autonomy of the community is acquired through strategically placing its actors into a deeper dependence to each other. If the work is not able to stand ten years' worth of environmental wearing, then the number of educated people available should make up for it. We are suggesting that these fragilities, those weak points, are implemented strategically to stir the structuration of the network of actors constitutive of a technical milieu.

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[3] Gestion de Terroir translate as ”terroir management”.

[4] This should not be mistaken for precarity, for it is evident that minimal knowledge access, know-hows valuing, and material accessibility are a requirement for fair and just living conditions.

The Grand Mosque of Djenné (Mali) was initially constructed before 1330 out of rammed earth. It was then found abandoned in the nineteenth century. After its reconstruction in 1907 by French colonialists, supposedly faithful to the original design (Leiris, 2007; Bourgeois, 1987), the Mosque has gained a great level of interest in its population. Nowadays, it has become a national symbol and mobilises most citizens for a now infamous yearly re-plastering[5] (Fig.01). Because sun-dried earthen plaster wears off rapidly given the environmental conditions of Djenné, it offers a yearly opportunity for care enactment with local resources (Brunet-Jailly, 2017). Because of its versatility and abundance, virtually any rammed earth could be used as a plaster; the local population sources its raw material from the floodplains of the river Bani, just outside Djenné. The *technical milieu is dependent in more than one way,* not only *of its external milieu* (planned wearing and frugal material availability), but also *of its internal milieu* – that is by means of manual craft transmission and the perpetuation of a yearly maintenance ritual.

Figure 1 - Yearly re-plastering of the Grand Mosque of Djenné (source: Peter Howard - African World Heritage Sites).

Reducing distance by embedding these technological fragilities is equivalent to setting intrinsic resources exploitation limits. In the French law that regulates the protected Salers cheese for instance, it is specified that it should be produced on the exploitation site where the milk originates, from a list of villages, between April 15th and November 15th, and when cows are eating field grass[6]. This is a regulation that makes it easy to identify the degree of locality of the Salers technical milieu: the materials are highly constrained, both in time and space; the method and tools are open, detailed and regulated by the law; the craft must be mostly manual. The mode of existence of Salers cheese as a technical object therefore protects all its resources while protecting locality by ensuring that the technical milieu is bounded to, for instance, not integrating an unsustainable level of automatism that would contribute to adding a distance between the internal and external milieus (Fig.02).

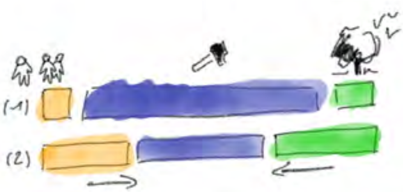

Figure 2 - Dynamics of milieus: case (2) illustrates an effort at balancing the influence of the internal and external milieus in relation to a technical one.

4. Fragile computation

This critical analysis of milieus draws attention to the importance of the networks of relationships through which technical objects exist. As discussed earlier, we suggest that overcoming the resources myopia fostered in the modern era and supporting a redevelopment of the relevance of the territory can be tuned, nudged, stirred by the method of strategically fragilising a technical milieu. Because silicon-based Information Technologies support a pinnacle of virtuality - exponentially gaining in coverage and

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[5] While the depiction of all the terroir and vernacular architecture can draw us towards romanticising them, the socio-economic externalities to the examples of technical milieus need to be acknowledged too as they can reflect a structural precarity (Brunet-Jailly, 2017).

[6] Décret du 14 mars 2000 modifié relatif à l'appellation d'origine contrôlée « Salers ».

integration (Cano *et al*., 2018) - we are using them as a departure point for a case-study: how could an information technology be conceived given an increase in material and human resources dependency for the system?

The idea that living complexes might act as computational devices was already under serious consideration in the 1960's. For example, Stafford Beer, the British cybernetician and management consultant, reported on his investigations in trying to establish input and output couplings with a variety of organisms including *Daphnia* and *Euglena*, culminating in a proposal to employ an entire pond ecosystem as a homeostatic control device (Beer, 1962; Pickering, 2006). Underpinning such a proposition was a keen understanding of the regulatory and symbiotic relationships that emerge in deeply coupled systems - an understanding also confirmed by Suzanne Simard, a Canadian researcher in forestry management and pioneer in the untangling of forest-fungi relationships. She qualifies the mycelial network of fungi as a "wood-wide-web" in forests, a chemical exchange system that sets plants in relation via fungi (Simard *et al*., 1997). Inspired by this idea, Fungal Architectures (FUNGAR), an EU H2020 Future and Emergent Technologies funded project, is developing ground-breaking methods for implementing a living fungal automaton at the architectural scale (Adamatzky *et al*., 2019). Building on years of research focused on understanding the computing behaviour of slime moulds (Adamatzky, 2016), it has been shown that the bioelectric activity of fungi varies depending on its environmental conditions. The invested fungi are ligninolytic saprotrophs. Their bioelectric activity is the result of enzymatic osmotic stress that is used to move previously decomposed nutrients through their cellular membrane for assimilation. As recent research shows, this activity is influenced by quick heat variation (proximity of an open flame, for instance), light intensity, and presence of chemicals (Adamatzky *et al*., 2021). Furthermore, the electrical spiking behaviour of fungi is being studied to interpret the various responses with acuteness. Reading the bioelectric activity is typically done by using a high-resolution ADC-24 data logger, with signal analysis performed algorithmically (Dehshibi & Adamatzky, 2021). This marks the limits of the vernacular techniques.
To increase the dependency to the natural milieu of this novel IT, on-site fungi baiting methods are being developed (Fig.03). In a given forest context, and after a preliminary mycological diagnostic, the intervention is composed of strategically located substrates that mediate the environmental conditions, such as moisture and heat, for fostering a controlled fungal colonisation of parts of the work. As suggested by the preliminary diagnostic, Fungal Architectures focuses on developing reproducible design methods rather than material, strain, or substrate specific designs. This is useful for challenging the dependence of the system to its natural milieu as the method incentivises designers to situate the technical decisions. It also aims to mitigate against the use of a limited selection of strains that might encourage human-led spread of non-native strains in a variety of environments, risking imbalance in local ecologies. The versatility of the investigated strains shows that the resort to wood and agricultural substrates, in all their geometric and chemical diversity, opens tremendous perspectives on composing complex yet vernacular artefacts based on mycelium. This opportunity encourages the development of methods that make use of local waste streams and available natural resources. In addition to the trigger, spiking, and signal-based computational behaviour analysis, the effect of architectural tectonics on the computational behaviour is being investigated. Because the bioelectric activity is influenced by its direct environment, the architectural design is being informed by environmental simulation to predict sun, wind, and rain exposition over time to stir the resulting fungal computational behaviour. The principal scaffold of the work is made using the resilient and ancient craft method of Kagome weaving (Ayres *et al*., 2021; Ayres *et al*., 2018). By integrating a holistic approach to the description and management of the fungi-based computation, Fungal Architectures aims at demonstrating that contemporary technologies and knowledge allow us to embrace complexity and serve a socio-environmental project.

Dead fungi also find a design interest in the present context: since their popularisation in 2009 with Philip Ross' Mycotecture Alpha installation, inert Mycelium-Based Composites (MBC) have attracted investigation in the fields of design and architecture; The Living's Hy-Fi intervention for MoMa PS1 in 2014 and MycoTree for the architecture biennale of Seoul in 2017 (Heisel *et al*., 2017) are demonstrative of the growing interest for this class of composite materials. Because ligninolytic fungi typically feed off agricultural waste, the production of inert products based on mycelium makes for a very affordable craft. Although fabrication methods necessitate an environmental cleanliness to prevent contamination of the substrate by undesired fungi and bacteria, kitchen-type methods are shared via a growing list of online resources ranging from social media groups to forums, blogs, and open repositories. Making an MBC is not unlike making bread, or cheese. The involvement of the craftsperson with the being, because it is fundamentally other and demanding (regarding contamination risks and its phenotypical expression), encourages a loss of the self into being attentive, caring for the understanding of the fungal lifestyle. The perspective that MBC offer in their vernacularity encourages a renewed understanding of the relationship that a technical object entertains with its craft and its sociological impact (Rigobello & Gaudillière-Jami, 2021).
We have previously discussed the importance of the craft in closing the gap between the user and the resource. In resorting to a material system that is principally vernacular, and with a distinct interest in the craft of MBC, Fungal Architectures exemplifies an on-going research led by a motivation to get the user closer to forest resources via unconventional Information Technologies. But the openness of the craft is

also instrumental, as the choice to develop an architecture that hosts an active saprophytic fungus introduces a peculiar fragility: decay. As years go by, a fungal architecture will decay rapidly and will require maintenance. Similarly to the Grand Mosque of Djenné, the architectural design needs to incorporate features to ease the maintenance, and the craft too must be, and is, accessible, affordable and versatile. Fungal Architectures qualifies as a terroir milieu and can hopefully inspire the development of architecture, design and engineering practices that are neither technophobic nor technicist, but of informed technics.

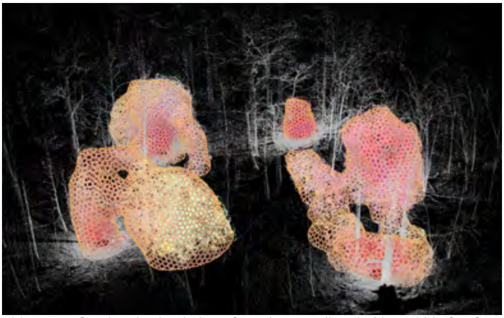

Figure 3 - Fungal Architecture. Stochastic simulation of on-site mycelium baiting within freeform kagome scaffolds.

5. Conclusion

After an epoch of apparent resource abundance, the acknowledgement of massive biodiversity depletion caused by globalised resource extraction (Turvey & Crees, 2019) should urgently lead to design (and engineering) practices that structurally encompass a new paradigm of resource economy in order to sustain the needs of the many and contribute to reducing global and local inequalities. This economy must consider not only the materials being used but be informed with Life-Cycle Assessment evaluations. Lessons from premodern and non-modern technical milieus such as in terroir bear much potential for inspiring designs for resilient communities and environments. The strict definition of the natural resources being mobilised by a given technical object or system with the terroir quality could contribute not only to protect the resources at stake but also to value local cultural identities and belongings - a postmodern project illustrated by recent research in xenodesign (Rigobello & Gaudillière-Jami, 2021) and urged by mobilisations such as the Black Lives Matter movement (Lebron, 2017). Making technical milieus less resilient in that they cannot replace a resource by another as the location matters, rendering them fragile, is to displace the centre of gravity towards environmental resilience. This effort, combined with an increase in the role of human actants within the technical milieu by strategically resorting to tacit knowledge sets via craft practices, contributes to fragilise further the technical milieu to the benefit of socio-environmental resilience. As shown through the example of terroir, a fittest novel formulation for the design practice we can afford would be *more-than-human centred*: economical in technics development, where the translation of a social activity into a nonhuman actant is evaluated beyond anthropocentric interests - as opposed to the modern progressivism philosophy; humble in scale as primarily focused on locality - that is an in-depth work taking up environmental complexity rather than simplistic and totalising; and situated, with an unavoidable characterisation of the mode of existence of the technical object with resources renewal. Economy, humility and situatedness; these qualities could form the ground for a design practice that challenges both a western relationship to techniques assuming an abundance of resources, and support the advent of affordable cosmotechnics (Hui, 2019) that support local identities and ontologies of nature. Furthermore, we see this terroir method as a grounded and productive implementation of advances in the understanding of socio-technical networks (Latour, 1990) and feminist studies (Haraway, 1991), hence its potential to help assessing more-than-human inequalities in leaving a capitalist relationship to technology for an informed cosmotechnic practice of design.

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F.2 Prototaxites stellaviatori: a fungal growth simulation model for Mycelium-Based Composites education in applied arts.

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Prototaxites stellaviatori: a fungal growth simulation model for Mycelium-Based Composites education in applied arts.

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ABSTRACT: The increasing experimental investigations of Mycelium-Based Composites (MBC) in design and architecture necessitate efforts from pedagogues to find ways to transmit knowledge and support regarding the guiding principles of mycology so as to empower students in their investigations and study. The adoption of MBC craft in arts and applied arts offers much potential in extending the space of material semiotics as it is often accompanied by several theoretical and systemic interests, such as the urge to adopt alternative ontologies of nature or implementing thoroughly sustainability in the economy. To support these critical reflections and exploring novel formal expressions we have developed a stochastic simulation model of fungal colonisation for design education and research in MBC. In this paper we present the conceptual and technical framework guiding the model's development with specific focus on its role as a pedagogical instrument. We report on its pedagogical impact based on a students survey and their productions.

1 INTRODUCTION

Mycelium-Based Composites (MBC) represent a class of materials that has only recently come to be used in design and engineering. Inspired by early XXth century method of fungal strain transfer by lignocellulosic solid-state fermentation (SSF) (Duggar 1905), the use of this craft in design was popularised amongst others thanks to the 2009 Mycotecture works of Philip Ross (Ross 2016). Since then, a wealth of design and architecture projects have been revealed, while the material semiotics of MBC is still young and being established. The cultivation principle of MBC relies on the saprotrophic lifestyle of ligninolytic fungi, or wood-rot activity, and consists in having wet lignocellulosic substrate colonised by a mycelium. Such mix is typically incubated at a temperature fit for optimal enzymatic activity for a few days, that is until the substrate has been colonised by the fungus according to the design intent. The colonisation speed and incubation duration vary depending on strains, substrates, and design objectives. This simple and frugal process of composite craft actually reflects a wide-ranging design space and complexity because it can be implemented with a wide variety of substrates, both in terms of chemical profiles and geometries, and a range of basidiomycota. Furthermore, the spread of MBC craft is often accompanied by theoretical and systemic interests, such as the urge to adopt alternative ontologies of nature, or implementing thoroughly sustainability and social justice in the economy, stakes that need fine integrating in applied arts educational programs.

The increasing experimental investigations of MBC in applied arts necessitates efforts from pedagogues to find ways to introduce students to the guiding principles of mycology. Such a knowledge set is complex in nature as it relies on an advanced biochemistry literacy that design students are not usually exposed to during the course of their studies. Thus, they must be able to grasp tacit knowledge — knowledge that cannot be formalised — of MBC crafting without tackling the full explicit knowledge set available in mycology. Furthermore, this craft must then be integrated with

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design expertise. Such design expertise is also relying on tacit knowledge and is taught accordingly by resorting to design pedagogy, in particular via studio works. The pedagogy presented here relies on the articulation of these two sets of tacit knowledge by having the students venture into physical experimentation with MBC. The pedagogical proposal has been assessed with the help of the Socialisation-Externalisation-Combination-Internalisation (SECI) model (Nonaka, Toyama, and Konno 2000).

In this paper we present the framework guiding the development of a simulation model, named *Prototaxites stellaviatori*, with specific focus on its role as a pedagogical instrument. Accordingly, we evaluate its impact based on student interviews and material evidence produced during an MBC teaching workshop in September 2020 at the Royal Danish Academy. The goal of this workshop was for students to quickly achieve design agency in material proposition (internal characteristics based on constituents and composition) and application (conventional architectural design proposition), to gain an appreciation of underlying mechanisms of the simulation model, to explore modes of manipulating the plugin to achieve design intent, and, through the process, to develop critical thinking on the design implications of designing and cultivating materials implying living non-humans. The evaluation of the pedagogical impact examines the architectural and technical strategies developed by the students as well as the semantics they resorted to in describing them, seeking to understand the knowledge conversions at play. Finally, we conclude on the students appropriation of the role of simulation models and MBC conceptual framework with regards to somatic tacit knowledge — knowledge relative to corporeal perceptions (Collins 2010). A teaching support document is made available in a public repository along with documentation of student explorations and design outcomes (Rigobello et al. 2021).

2 TEACHING CONTEXT

2.1 *Pedagogical context*

Numerous post-structuralist and new materialist writings have been advocating for the adoption of a relational Kantian aesthetics based on affect as a counter discourse to modernist rationalism (Rigobello and Gaudillière-Jami 2021; Lowenhaupt Tsing 2015). While learning MBC cultivation methods is part of our workshop syllabus, it is critical that fungi is not only presented as a mere technique but in a manner that allows students to develop their tacit knowledge of the fungal life with the full extent of their corporeal sensibilities. Building upon an applied arts pedagogy that recognises the centrality of synthesis, analogy, and craft practice in the design expertise (Schön 1985), our drive towards conveying theoretical controversies finds a favourable ground, and is expected to echo and augment prior cenesthesic intuition in students — the blend of various bodily sensations producing a tacit awareness of being in a particular physical condition. As MBC cultivation is being democratised with commercially available kits for individuals along with a profusion of do-it-yourself workshops, and thanks to the process being frugal in not requiring expensive equipment or advanced skills to reach a satisfactory result, we can witness that a detailed understanding of the biochemical processes at play is not mandatory for using MBC as a design medium, thereby rendering it very accessible and inclusive.

The architectural design master programme Computation in Architecture offers a computational design and digital fabrication-oriented program hosted at the Royal Danish Academy. At the beginning of each academic year, both fourth and fifth year students are grouped for a series of introductory workshops designed to immerse them in materially-focused computational design logics that are thematically linked to active research projects led by the Centre for Information Technology and Architecture (CITA). Through this research-led teaching, students are exposed to a coherent set of questions, theoretical positions, concepts, methodologies and state-of-the-art materially oriented design and production workflows for them to anchor their subsequent individual semester and thesis projects. Fourth year students rarely have prior experience with computational design or biodesign. The workshop forming the focus for this paper, consisted of a preliminary two-week session called *Shaping Growth: composing material heterogeneity* that had the following objectives: producing a cast for forming and growing a mycelium-based composite, composing the substrate such as combining bulk material together with specifically orientated fibres, being introduced to methods of sensing and real-time monitoring of living materials, together with data-collection and querying, engaging in digital modelling of physical artefacts and be shown how to situate them within an extrinsic modelling environment allowing to generate

artefact performance data from environmental simulation. A second two-week session called *Liquid Modelling: navigation beyond vision* was held directly after the previous one. Its objectives are formulated as learning hypothesis in section 3.3.
A supporting document synthesising the fundamentals of MBC craft and of the functioning of the *P. stellaviatori* plugin was provided to the students at the beginning of the Liquid Modelling workshop. This document is made available in the same public repository as the documentation of student work and results (Rigobello et al. 2021). The workshop sessions embedded a day of lectures and tutorials each, the remaining of the time was dedicated to group work.

2.2 *The Rhinoceros 3D - Grasshopper environment*

Grasshopper is a visual programming environment within the Rhinoceros modeller. It uses visual programming, a particularly popular method of learning programming for novices, as studies of such techniques as used not only in the computational field but also elsewhere point out (Celani and Vaz 2012). Grasshopper is a didactic environment thanks to its online supporting community. Many observers associate this environment with what Eric Raymond calls a *bazaar*: a software environment that emerges through contributions from a larger, open community. Raymond crafted the term in opposition to *cathedrals* traditional software environments, referring to larger applications developed by a small group in a closed environment (Raymond 2001). Because visual programming facilitates abstraction and the acquisition of logic, its learning curve is meant to be less steep than for command line programming. However, the curve quickly reaches a plateau, trapping users in a narrower range of possibilities. Visual programming therefore poses a challenge in terms of upgrading users' skills (Celani and Vaz 2012; Aish and Mendoza 2016).
Despite its limits, Grasshopper was selected for the implementation of the *P. stellaviatori* model as it allows for articulating with other pedagogical methods and therefore enables the coexistence of a variety of learning strategies. One of the main issues in education in the computational field in architecture is indeed the need for the combination of several learning goals across multiple fields. While the students are expected to learn programming skills, they are also primarily in a design course and must be able to develop skills relating to the acquisition of architectural expertise. In our case, the students are expected to experiment with mycelium and learn cultivation processes along with developing critical thinking regarding simulation activities in design. We thus consider Grasshopper to provide a particularly suited platform for us to develop a multi-scalar pedagogy that relies on three pillars: programming, designing, and cultivating. We resort to Grasshopper to enable the interfacing between the *P. stellaviatori* model and solar radiation simulation, and to articulate the reinforcement of the connection between programming and physical experimentation to support the acquisition of somatic tacit knowledge.

3 METHODS

3.1 *The P. stellaviatori model*

A fungal growth simulation model was designed as the *P. stellaviatori* Grasshopper plugin. Fungal colonisation is driven by a wealth of factors including moisture qualities and dynamics in the substrate, the fitness of the substrate chemical profile to fungal species enzyme array, the aeration of the substrate during fermentation, the UV stress, the presence of other microorganisms, and the protocol care by the producing person. The simulation model focuses on integrating evaporation and capillarity as prime vectors for predicting fungal colonisation, because the presence of capillary water in SSF substrate is a prerequisite to allow extracellular transport of metabolites in fungi, and to establish osmotic potential for allowing turgor pressure and subsequent hyphal growth (Money 1995). In timber, moisture can exist as bound water within cell walls below fibre saturation point (FSP), free liquid water in cell cavities above FSP, and water vapour. Water presence in wood cell walls and cell voids (over-hygroscopic range) is the fittest configuration for optimal fungal growth in lignocellulosic SSF. We can also note that fungal decay by oxidation produces water along with CO_2, but is not a phenomena that is considered for modelling in this context. Similarly, the freeing of bound water under the action of decay that influences the moisture content as substrate dry mass loss is occurring if the system is closed, is not modelled. Lastly, the effect of water activity upon turgidity and enzymatic activity is not modelled. The simulation model can be used with hypothesising these non-modelled experimental factors as fixed, and can be used to explore the remaining of the design space allowed with its parameters (input datasets and tuning parameters).

The simulation is based on a spatial discretisation in the form of a volumetric point-cloud representing the volume of a substrate. The moisture dynamics simulation model comprises two sets of phenomena: evaporation and capillarity. The effects of fungal decay over moisture release from the substrate by cell wall decomposition, and carbohydrates oxidation, are not modelled. Water activity is not present either, but can be considered at calibration stage. The model resolution aims at being set at the scale of 1 – 5 mm distance between points of the cloud, and although it represents fungal growth as a graph, it does not aim at modelling hyphae networks but regions and patterns of colonisation. The macro fungal growth that is simulated represents the mycelial exploration of a substrate. The model takes four datasets as input: an initial moisture distribution map, a thermal map, the position of fibres within the volume, and the position of mycelium seed points. At iteration n, evaporation is simulated such that the moisture contained at point p is being partially transferred to its neighbours within an influence radius, in an eccentric manner to the favour of the neighbouring points situated above p, and depending on the warmth level at p. Capillarity is simulated in a similar way, but instead of considering neighbouring points it refers to clusters of points within a fibre and distributes moisture from p situated within a fibre, depending on its warmth level, within an influence radius, and in any orientation of the fibre (gravity is neglected). From a set of seed points within the point cloud, mycelial colonisation is simulated as a graph that follows the iterations of the moisture dynamics with a minimum moisture level parameter allowed for the growth, and a maximum branching level.

The model as defined is subjected to uncertainty: evaporation and capillarity simulations are meant to be accurate enough to be calibrated and instrumentalised for design purposes, but depend on complex models that are only approximated here. For this reason, and to convey the conceptual framework mentioned in section 2.1, this model was made stochastic. Therefore, each data point in the three initial datasets (warmth map, moisture map, and fibres parsing) has a corresponding probability. The determination of these are made manually by the user and are related to confidence in each input of the experimental setup. For instance, a user can set a high temperature on the external boundary of the experimental setup with a maximum confidence level of 1.0 as they can measure the value easily, and then set a lower temperature level at the centre of the volume of the experiment with a low confidence level as they may not be able to measure this temperature. While the simulation is being solved, the probabilities are being solved too as the intersection of the events at each point of the model.

3.2 *Mycelium-Based Composites methods*

The substrates and scaffolds materials were recommended to be prepared at 40 % MC with mineralized water, and sterilised at 117 ºC for 30 min. The substrates were inoculated in a variety of manners by the students. They were advised to mix the wet substrates with 16 wt% *Ganoderma lucidum* spawn (M9726, Mycelia BVBA, Nevele, Belgium) and to incubate them in PP filtered bags for 7 days at 25 ºC in the dark when relevant. The fungal species was selected for its widest carbohydrate-active enzymes array (CAZymes) (Zhou et al. 2018; Chen et al. 2012) and its colonisation rate, thus allowing students to reach quick conclusions with a high cultivation success rate. Among the most used substrates during the workshop were European beech wood (*Fagus sylvatica*), common reed fibres (*Phragmites australis*), and rattan fibres (*Calamus manan*). Students used a variety of supplementary materials such as glass, copper, or hessian (Rigobello et al. 2021).

3.3 *Evaluation method*

To assess whether the pedagogical goals have been met in the workshop, we have formulated two pedagogical hypotheses. They read as follows: a) *the teaching environment enables the students to articulate experiments with simulation use to support critical thinking development on the role of simulation models in architectural design*, b) *the teaching environment supports the students appropriation of the MBC conceptual framework*. In order to evaluate the pedagogical set-up and its consequences on the learning process of the students, interviews were conducted with each group of students. They happened on September 29*th* 2020, a day before students project presentation. The survey questions read as follows:

- What is your background prior to taking part in the Computation in Architecture master program? Did you have any knowledge of working with living organisms before this workshop?
- What are you working on during this workshop?
- Why have you chosen to explore this?
- Can you describe the experiments and simulations you have done and are planning?
- How have you been working with the simulations on one hand and the physical experiments on the other? How do you link them?
- How are the tools you were given helping you investigate the architectural potential of mycelium?
- What about the sensing protocol? Beyond the measures, do you have other sources of information on what is going on with the mycelium?
- How did you use the five key notions shown at the beginning of the workshop?
- What have you learned about working with mycelium?
- Do you think you can or will use this knowledge on other occasions? What architectural possibilities do you envision?

Since the 1990s, a great number of studies on architectural learning stemming from the field of design studies have been accompanied by research aiming at identifying systematic design methods in order to establish teaching models (for a review, see Demirbas¸ and Demirkan 2003). While only some of the existing studies identify architectural knowledge directly as tacit knowledge (Kruchten et al. 2005; Uluoğlu 2000), a great number identify the tacit dimension of the knowledge mobilised in architectural design, although they do not refer to it in this term, preferring notions such as *intention*, *intuition*, *implicit reasoning* or *empiric method*. To the knowledge of the authors, none of these studies assessed the development of tacit knowledge in an architectural teaching environment, despite the fact that diverse models are employed in existing studies to assess various aspects of the mobilised knowledge and methods in architectural training. In order to understand the knowledge transfer at play in our workshop, we have selected the SECI conversions model, which enables the assessment of tacit knowledge transfer (Nonaka, Toyama, and Konno 2000). This model has been conceived from the Japanese notion of *Ba*, a word that translates as “place”. *Ba* designates a place and time collectively shared, and includes the mental space that is shared when a group gets together. This shared mental area provides a space for knowledge to be exchanged and the conditions of this exchange — and conversion — are what the SECI model assesses. It allows for the identification of four modes of conversion: *socialisation*, enabling a tacit-tacit knowledge conversion, and referring to moments of observation and imitation; *externalisation*, enabling a tacit-explicit knowledge conversion, and referring to explicating knowledge in writing or drawing, for instance; *combination*, enabling an explicit-explicit knowledge conversion, and referring to the use of existing explicit formats in order to produce new explicit ones; and *internalisation*, enabling an explicit-tacit knowledge conversion, and referring to the acquisition of tacit knowledge through the manipulation of explicit formats. The SECI model, named after those four conversions modes, envisions these as successive and has therefore been criticised for being too linear. We thus use the model as an analytical tool, not using the conversion modes in a sequence but independently: these modes have been coexisting in the workshop context.

4 EVALUATION

4.1 *Interviews*

Among the most prominent ideas that were discussed, and because it was a focus across the two parts of the workshop, the establishment of an iterative process between experiential learning and simulation exploration was a leading interest. The discussions oscillated between students reporting on the successful integration of experimental learning into the simulation (as per growth rate tuning, environmental influence calibration, capillary action tuning, evaporation rate and reach), with group 3 having confidently discussed their success at calibrating the simulation model, and four out of seven groups having brought up supplementary experimental series being necessary to them for improving the predictability of the simulation outcomes. We read this as a necessary development of a critical thinking as per the role of the model as an instrument, and a relevant feedback as the students were both in their first introduction stage with mycelium craft, and expected to appreciate a non simplistic relationship to fungi. Five groups discussed the resort to simulation as a means to probe architectural design as grounded into a cultivation practice, and used the simulation to imagine constructive methodologies for load bearing structures. With use of solar radiation analysis within the design workflow, all groups have developed scenarios utilising solar warmth and its effect on evaporation to design either hand handleable modules that can be assembled after cultivation, or on-site cultivation principles. As the students developed their tacit understanding of the craft, they productively criticised the lab cultivation conditions that systematically use substrate sterilisation for instance. Four groups have specifically commented on the limits of the simulation model and the constructive unpredictability in an on-site cultivation scenario because it would involve accounting for microbial coexistence. Beyond scaling, four groups have reported making use of the simulation model as a way to probe complex designs based on elementary experiments they conducted. A group reported their interest for using it to build a finer understanding of the colonisation at play with a volume of substrate, beyond vision, while group 6 documented experiments specifically testing a moisture-based powder bed printing lookalike method. Group 2 also ventured down this path, by rotating a composite during its cultivation so as to change the face being exposed to the sun and successfully influenced the moisture spread. Other groups have been using the simulation not to assess more complex craft methods, but as a means to establish hypothesis and evaluate experimental setups that they would not have had the time to perform.

Across groups, the feedback regarding compressive performance appreciation was heterogeneous: two groups reported doubts as per the state-of-the-art mechanical performances of the composites, and two other groups expressed their surprise regarding the good stiffness of MBC. This perception reflects the architectural potential that the students envisioned. Some groups praised the more-than-human collaboration with a mycelium, mentioning resulting forms that we would not intuitively trust as humans, making it an opportunity to challenge pre-existing ideas of comfort, pleasure and sterility. The smell of MBC was flagged as intense by a group, opening a productive conversation that started from the need to cancel it and went on to refer to the comforting perfume of wood: ”Do we grow to enjoy the smell of wood, or do we instinctively appreciate it?”. For some, it was a potential unpredictability combined with the exploration that was motivating. A student interestingly reported that the craft - the process - was similar to doing a renovation project that necessitates the rediscovery of obsolete constructive methodologies and materials. While acknowledging that MBC can be controlled and be made stiff and resilient, notably with the introduction of glass and cotton in the substrate, some students advocated for the necessity of the craftsperson to also be sensitive to the colonisation and mycelial expression for allowing the full expression of the craft. Echoing the smell controversy, the same group went on with the idea of comfort to envision fungal architectures as uterus-like, where elements are swollen, soft, and reminding of embryo state.

Throughout the interviews we read a good appropriation of critical thinking on the role of simulation models. Overall, the introduction of the stochastic layer to the simulation model added a third level of learning to the workshop in addition to MBC craft introduction and getting introduced to computational design and its functioning. No explicit mention was made of this aspect in the interviews, and although each group made some sense of it in their submissions it is obvious that the combined complexity of the three key notions was detrimental to the students appropriation of the stochastic functioning. Its contribution is hardly distinguishable from other factors of the pedagogical environment, but the considerable involvement in crafting by experimenting creatively and the resulting conceptual appropriation beyond considering MBC as sole materials can surely be credited for a part to having made uncertainty a defining trait of the model.

4.2 *SECI conversions*

In order to discuss the insights obtained through resorting to the SECI model, we discuss the events of the workshop through the review of the knowledge carriers in presence as well as through the examination of the activities that took place. Both then serve as a basis to establish which conversions happened during the workshop and whether their presence or absence yielded the expected results. Written carriers include the teaching support document, and the presentations produced by the students as a final deliverable (Rigobello et al. 2021) Knowledge carriers also include the physical objects crafted by the students. We consider these objects to most prominently carry a different form of knowledge, as it is a trace of the craft somatic knowledge acquired by the student. In particular, the attention to materiality that they demonstrate through the combination of materials in the substrate and its colonisation by the mycelium acts as a trace of that acquisition. In that sense, while written knowledge carriers testify to the existence of externalisation and combination modes in the workshop, the objects testify to the existence of socialisation. Furthermore, the workshop as a learning space has been the opportunity for other occurrences of socialisation as well as of internalisation, both through the observation and imitation of model manipulation and of craft practices. The interviews confirm that these latter conversions, happening in the workshop space, have played their role in the acquisition of new knowledge by the students. The presence of the four SECI conversions and the confirmation through interviews and documentation of their impact on knowledge acquisition thus validates the pedagogical set-up by providing insights on how explicit and tacit knowledge have been transmitted. The workshop as a teaching format is a fit environment for architecture students to acquire a relevant knowledge set thanks to the modalities tackling both explicit and tacit knowledge acquisition. This is confirmed by assessing the materials produced by the students with regards to the two hypotheses we formulated previously. The sheer diversity of objects and experiments that students crafted are a testimony to this. So is the student's visual production, which mobilises the technical information provided in writing as well as exploring various modes of representation provided through the *P. stellaviatori* tutorials. The documentation produced by the students highlights their attention to material choices, shown carefully through composition detailing and texture focuses. It also highlights their appropriation of the various factors influencing mycelium colonisation which is reflected in the architectural proposals.

5 CONCLUSIONS

In this article, we have presented the conceptual framework for developing the *P. stellaviatori* simulation model as a pedagogical instrument. We have reported on the pedagogical theoretical framework we adopted for evaluating the effectiveness of the simulation model as part of the proposed teaching environment, and have presented a comprehensive analysis of student group interviews specifically informed by the SECI model. Craftspersonship was a central theme in the interviews, be it indirectly through reports on somatic perception (i.e. smell for instance), or directly in the constructive methodologies of student proposals and their instrumentalisation of experiments regarding the simulation. With the first half of the workshop having been focused on the MBC craft initiation, the craft nudge allowed by the simulation model is identifiable through the moisture focus that students developed in their tests. All groups have adopted this focus, and its integration with solar radiation analysis led part of the students to experiment outside of lab conditions to test context hypothesis. This had a productive influence over their architectural visions, and across the seven groups a diversity of grounded constructive methodologies are presented in the supplementary materials (Rigobello et al. 2021). Finally, while we acknowledge the density of the expected learning in this constrained workshop period, our learning hypotheses have been corroborated; they regarded the development of a critical thinking as per the role of simulation models in design activities, and the acquisition of a somatic tacit knowledge for a MBC conceptual framework. This leads us to conclude on the effectiveness of the use of this simulation model as a pedagogical support embedding traits that facilitate the setting of an applied and conceptually situated knowledge, that is within the larger set of used teaching media.

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F.3 Designing the Gross. In search for social inclusion.

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Designing the Gross. In search for social inclusion.

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Abstract | Numerous researches dedicated to the gross exist in psychology, focusing notably on the sense of otherness behind this reaction of disgust and fascination. Furthermore, many grotesque works of art have been achieved, in particular in the XXth and XXIth centuries, in attempts to challenge prevailing social and moral models.
Contemporary design practices, inheriting from a universalist desire of modernism, presuppose a single, unitary identity built according to dominant social constructs. They are nonetheless questioned by the recent emergence of speculative design projects employing materials traditionally deemed disgusting.
The present research examines from a material semiotics stand the gross at play in queer & feminist art and in recent speculative design works. As these works show exemplary potential in questioning moral *a priori*, this research sets them in discussion with a design research program conducted by NGO design studio thr34d5 on kombucha pellicles and their craft. The theoretical and experimental analysis presented in this research form an understanding of the role that material semiotics combined with the gross can endorse to leverage design in matters of social inclusion and impact.

KEYWORDS | GROSS, OTHERNESS, KOMBUCHA, DESIGN THEORY, MATERIAL SEMIOTICS.

1. Introduction

Wet, slimy, sticky, mouldy, rotten, spoiled, decaying, smelly… Gross. The construction of taste as what is commonly acceptable is built throughout centuries of collective memories. In present times of peak in social inequalities and societal need to recognise and give agency to situated communities, it is this shared memory that is urgent to understand and seize methodologically. The modernist globalisation of consumption habits shapes most of our societies into clean and smooth realms, with a universalist goal of standardizing our ways of being, our habits, our instincts. This universalisation that pervades in the design of everyday objects and services keeps pushing further away identities that do not fit within the norms by instrumentalizing moral into aesthetic forms and technical functions. This resorts into accidental or strategic suffusing of new designs with a semiotics of dominant models – principally for dominant classes privileges continuation, whichever they are.

Gross phenomena nevertheless resist, but rendered marginalized as they illustrate behaviours that defy dominance. Gross as considered here is the combination of disgust and fascination, defined respectively as the "*experience of intense physical dislike for an object or a situation*" and the "*urge to explore, investigate, or to understand something*" (Fokkinga & Desmet, 2013). It is in itself the subject of many studies in the field of psychology and has become an activist symbol in art and emerging xenodesign[1] practices as a way to question the construction of identities. Beyond fetishism, mobilizing this emotion is a call for otherness. Accepting the other, be it a person that endorses a different culture or a non-human being, is to agree to confront ourselves to what is uncommon, to what is beyond frontiers and control. The semiotics behind the works resorting to the gross contribute to form a relational aesthetic opening a discussion on the role of collaboration in the design methodology, a defining quality at stake in this study.

The present paper introduces a design research program conducted by NGO design studio thr34d5 on kombucha pellicles. This microbial community was chosen as it is both alive and unsettling in its materiality, and presents the benefit of having a low cultural anchorage. From the perspective of material semiotics, this study discusses the holistic sensorial approach of grossness in the making of kombucha pellicles and presents the potential that lies in this emotion and semiotics focused method for envisioning a richer, more inclusive and impactful practice of design.

[1] Xenodesign is a term constructed by using the *xeno* prefix, which sits in the context of speculative realism related theories. It aims at describing techniques of alienation as productive ways to think about the unknown (Shmeer, 2019).

2. Gross is not grotesque[2]

2.1 Transgressivity as a project

"*Disgust is an ideological response to something that is offensive to the self because of its nature or origin*" (Rozin *et al.*, 2008). While neurological studies aim at evaluating the innate part at play in our disgust reactions (Moll *et al.*, 2005, Toronchuk & Ellis, 2007), a wide literature investigates its mechanisms of acquisition. While often criticized for their static analysis that does not taking into account the evolutions of disgust in a given culture or individual across time, and the possible accustoming to initially revulsing things (Lupton, 1996), Mary Douglas and Pierre Bourdieu have demonstrated that the sense of disgust is a social construct (Bourdieu, 1979; Douglas, 1966). While Bourdieu offer an analysis of the relationship between class, habitus and taste, Douglas has shown that not only does the sense of disgust vary from a cultural context to another, but it is a tool for maintaining social systems and boundaries – a remembrance of biopolitics in Foucault (1972). "*Feelings of disgust function to dignify humanity by allowing humans to put themselves above the animals that are deemed as inferior*" (Goldenberg *et al.*, 2001). Disgust crystallises the will to distance oneself from creatureliness; and we witness how this quality is being instrumentalised with intent, for instance in attempts at controlling one's body shape (Angyal, 1941, Rozin *et al.*, 2008., Goldenberg *et al.*, 2001). If we are to extend the argument, this distanciation is not only from what is animal, but from what is other, made foreign. The mechanics of it justify a rejection of what is not deemed moral to the profit of what is, as seen in resources exploitation and in formation of castes. It ties back to a larger phenomenon of exclusion, discussed in Foucault as instrumentalized in the production of truth and subjects' control. A central matter since his early works, Foucault analysed one of the apparatuses at play as the partaking of mental disorders in the process of legitimising discourses, proceeding to the exclusion of subjects deemed mentally ill to moral margins.

The elaborated body of work provided by Foucault set in perspective with our present object of inquiry, the gross, highlights the process of construction of our social identities through alterity. In an ever-expanding world of multi-cultural encounters, the project of a universal western moral meets raising defences for the recognition of situated identities beyond fringes or spectacles. Design at large, as the discipline of the construction of the synthetic world – not to say cosmos – can and need to seize the opportunity of recognizing its importance in the mediation of everyday interactions, thus becoming instrumental in building a togetherness around this alterity.

[2] "Grotesque" is a French word that designates both the baroque mobilization of the gross in art, and a ludicrous act.

2.2 Mobilizing the gross

The disgust is presented in psychology as a form of construction of our societies; how do these unmanageable phenomena perform in the construction of the self of the social life? A major part of the construction of our identities is mediated through our senses (Friedman, 2016). There are many forms of solicitation through the senses, of models that we have tested, inherited or accepted as part of our individual cosmos. Challenging those models helps us to better contour ourselves by sharing experiences of the construction of reality. Grossness represents then a negative space - it is part of this photographic impression of our social constructs for what is other, what we have been taught to fear and question, what does not *belong*. In times of uncertainty and divide among cultures, confronting fears is a movement against the grain, against borders to identify liminal spaces. To question those borders is to accept for one to change themselves, to challenge their identities. The fascination at play towards disgust is a call to embrace and adopt the fear of the unknown to try evolving our understanding or reality construction. We are other to others; everyone is a stranger in a quest to become object of others' fascination - to be adopted as part of a social group. Designing with the gross is one way to open up a shared platform, a public space for collective identities to advent.

Throughout history, arts heavily relied on triggering this mixed emotion for the viewer to put dominant models into question and enquire on the definition of identity. Tackling unravelling emotions calls for a discussion about censorship. As a globalising universalist moral censorship, violent and radically unifying in the form of modernism, some artists engaged in pushing the barriers of what is deemed acceptable. Questioning axioms of dominant cosmogonies, Duchamp's *Paysage Fautif* (1946), Serrano's *Piss Christ* (1987), Quinn's *Self* (1991) or Delvoye's *Cloaca* (2000), form an ensemble of provocations stirred by using "abject media" (Kristeva, 1980): semen, urine, blood, faeces. The mechanism of these works of art relies on the fascination from these various contexts - sometimes religious, always linked to our collective identities - and questions the conventional material semiotics by a raw transgression (Law, 2008).

Challenging the legitimacy of dominant models with the help of disgust and fascination is an antimodernist stand. The hierarchical, controlling relationship to nature is predominantly questioned by resorting to the gross; among most famous contemporary artists, Damien Hirst or Pierre Huyghe regularly mobilize living entities, including some usually perceived as unsettling. Picture venomous snails, flies, bacteria, and even cancerous cells. Huyghe articulates gross, life and otherness together in his pieces - notably in *After ALife Ahead* (2017), *Colony Collapse* (2012) or *Untilled* (2013). While he advocates for his installations to be self-organized systems growing on their own, "indifferent to the public", the fragile equilibrium reached between artificial and natural components in his works acts as a call for reassessment of our place in the world for the viewer (Russeth, 2017). It is with rotting flesh and food installations such as *A Thousand Years* (1990), *Mother and Child* (1993) *or Let's Eat Outdoor Today* (1991) that Hirst develops a reflection on "*human attempts at isolating and* *retrieving horror from our lives*" (Ghenasia, 2017). His *memento mori* recall the association demonstrated between death awareness and the feeling of disgust (Goldenberg, 2001), as he underlines the violence of our relationship to flesh, the severance of our relationship to nature and our inability to relinquish control.

Since the 1960s corporeal fluids and normatively gross body details have been reappropriated by queer and feminists' movements to protest universalist injunctions by political oppression and physical standards of domination. Feminist art, with works such as the *Read My Lips* campaign (Zoe Leonard and the collective Gang, 1992) or the especially extreme *Rhythm 0* by Marina Abramovic (1974), exposes the control of women bodies and question the role they are conferred in patriarchal societies through aesthetics of shock (Martinique, 2016). Such works helped perceive body norms as particularly strong mandates in contemporary societies, thanks to radical meanings they suggest by instrumentalising materiality. Contemporarily to feminist art, since the 1920s a queer art has been emerging and recognized for its artistic value, but also for the collective conditions of its emergence - challenging our perception of the artist as a masculine isolated genius for art as a social activity (Alfonsi, 2019). Queer art is characterized by its visceral challenging of rules aiming at normalizing identities. This current in art is activist, but beyond a political banner, it engages in making visible public claims for a revised humanism. Questioning one's own being through themes such as hybridity, mutations of the body, and the self, queer artists are illustrative of a liquid modernity (Bauman, 2000) and of a societal need to leave space for a variety of identities and cultures. Mobilizing disgust in art thus shifts from a grotesque staging of flesh & fluids to more subtle issues of identity recognition and discourses acknowledgement in the tradition of humanism critique.

2.3 Liquifying design

Grotesque in design has been more scarcely used and examined, with the exception of (Fokkinga & Desmet, 2013) and of children toys (Livingston, 2019). Speculative design increasingly relies on provoking a mild feeling of disgust, in a similar fashion to the works of art previously cited, in order to trigger a long term reflection on the contemporary hybridization of identities; some illustrative works are: Sinae Kim's *Urine Ware* bowl designs shaped as urinary bladder and using urine as a glazing material, Studio Basse Stittgen's *blood related* series of objects made entirely out of human blood - some pieces of the series even being made out of HIV positive persons' blood -, Studio Nienke Hoogvliet's *Waterschatten* lighting, table and bowls made of used toilet paper. In these three cases, the sense of disgust coming from the materials is a reminiscence of abject media as used by artists. As an answer to the analytical culture infused in modernist design, we find again in those illustrative works a mobilization of materials that are morally unacceptable. Even though the productions are delicate and aesthetic, they embrace disgust and make it fascinating.

In designs are encoded contemporary beliefs, contemporary cosmogonies; Slavoj Žižek illustrates this concerns through the phenomenology of toilets: "*We have such a multitude of* *the toilet types because there is a traumatic excess which each of them tries to accommodate - according to Lacan, one of the features which distinguishes man from animals is precisely that, with humans, the disposal of shit becomes a problem.*" (Žižek, 1997). Design – as craftsmanship and in its lineage – has become a principal vector for the mediation of phenomena through technics, and its practice has recently evolved as a new form of crafts of affordances, an engineering of virtual and/or material interfaces to phenomena. Design is never neutral but an artefact of an ideology, of a sociotechnical system it belongs to. As design was born with fashion-led planned obsolescence, we understand that the conception of objects and services can be driven from semiotic concerns that go further than optimizing their supply chain and interfaces for financial purposes and competitive risk mitigation (by green washing, for instance) but need be appropriated to build a rich and inclusive practice. Thinking design practices as neither enslaved to financial purposes nor elitist in their speculative qualities, but meant to positively impact situated communities and empowering to users, it represents a massive negative space around current practices.

Xenodesign is an emerging critical practice inheriting from speculative realism schools of thought. Explorations in this field focuses on decomposing the genetics of design systems so to identify their principal qualities and hack them, by replacing marble by coagulated blood for instance. Xenodesign artefacts situate in the context of hyperobjects, challenging socio-cultural events such as sustainability, AIDS, or gender inequalities (Morton, 2013). If design is an act of negotiation (Rigobello & Gaudillière, 2019), then xenodesign is an activist act towards social inclusion. It is an answer to aforementioned liquid modernity, which characterises by an increase in the rate of change of contemporary institutions and beliefs, which is to say that are subjected to an ever-decreasing lifespan. They therefore cannot "solidify", leaving individuals to have to constantly reassess their environments, decide and act in a fragmented way. This indeterminacy and unpredictability reflect on our ways to live and act upon the world, and ultimately on our ways of being. The western universalist project that is conveyed by capitalist led designs is thus being questioned by xenodesign practices, hence highlighting the need for a recognition of multi-cultural coexistence through what mediates our social life: objects and services.

Similarly to the critic that Alfonsi (2019) builds on art history analytical traditions biased by the masculine idea of the isolated genius artist, xenodesign practices often find an antimodern alternative is its modes of production. Collaboration in craft is found in queer theories, and in inherited artistic practices as with Enzo Mari and Sol Lewitt, or in Tom Sachs and Thomas Hirschhorn, to name a few. This change in modes of production is also nested within Donna Haraway idea of companionship, which drives xenodesigners into a struggle to build pathways to a co-dependence with non-human beings, reminding of the search for extended collaboration. Such a methodology we recognize as radically inclusive: one that is offering space for an expression of the identities, emergent, community centric (Rigobello & Gaudillière, 2019). Becoming less and less viscous, a xenodesign is emerging under collaborative and vindictive forms. In fact, the millennial or centennial traditions we previously relied upon for a social belonging, such as sharing a family recipe that is passed on over generations, tend to fade away as they are being partially replaced by faster changing values systems embedded in everyday designs. Novel craft practices as Do-It-Yourself and Do-It-With-Others in the domains of electronics, biohacking, mechanics and woodworking as seen in fabrication third places demonstrate means of production that valorise the social life of designs through open practices. On the contrary, examples of instrumentalization of these social ways of being are found in major social media businesses, or open research platforms relying on a needed belonging to contribute to a collective constructive project.

Design represents a tremendous yet dormant potential of methodological improvements that can foster a sense of togetherness in alterity, that is by promoting a “repository-forking” strategy allowing for the co-existence of multiple yet situated genetic and memetic evolutions for objects and services. On another level, and as is manifested in open-source platforms, efforts need to be put towards favouring a holistic approach to design supporting social inclusion and an increase in knowledge transmission affordability; in haptics belongs a relay to enrich the wideness of points of entry to a shared idea, as illustrated by the gross.

3. A craft of all senses

3.1 Kombucha as a canvas

Determined to advance the comprehension of the social inclusion potential lying in design disciplines, NGO design studio thr34d5 decided to test out hypotheses about the gross in the presence of a complex of micro-organisms and/or material system that have a low cultural baggage: kombucha.

Symbiotic Cultures Of Bacteria and Yeast (SCOBY) are co-existing communities composed of microorganisms – kombucha is part of this group. Kombucha is composed of a zoogleal mat grown aerobically by *Komagataeibacter xylinum* bacteria from a sweet tea solution (designated as “the pellicle” here), which protects from contamination *Zygosaccharomyces* yeasts feasting in the sweet tea solution, while using ethanol produced by the latter to produce the mat. The fermented solution eventually turns into a sought-after probiotic beverage for humans. The fibres of the pellicle are composed of bacterial cellulose, which is about a hundred folds thinner than the fibres found in plant cellulose (Schurz, 1980), making it a competitive material to synthetic polymers. Kombucha has a mythical origin. It traces back in Central Asia (eventually from Manchuria, China), and has apparently spread from this region over a period of approximately two thousand years.

The kombucha pellicle can be dried out, obtaining thus a material similar to leather, textured, that can be used in a comparable fashion to craft objects. Several designers or engineers have worked with this material, making garments (Suzanne Lee, Scobytec), packaging (Elena Amato, MakeGrowLab) or small leather goods (Echocella). In continuity with peer-to-peer sharing practices sustained for many years around the preparation of the kombucha beverage, designers such as Suzanne Lee have developed their work under Creative Commons licences and shared their expertise in open-source organizations such as biolabs. The information shared on kombucha pellicle and its production focuses nonetheless on the growth recipes rather than on other aspects of the process for instance, drying and post-treatment methods. Furthermore, many companies have recently formed selling the kombucha beverage, and product designers are launching commercial initiatives based on kombucha leather goods. A difficulty subsists to industrialize the production of kombucha pellicle: while designers have conducted research on the material, its production process, possible textures and colours, and crafted objects, there are little replicable initiatives due to *K. xylinum* phenotypic expression and variations from strains' origins in yeasts and bacteria ecologies. Internal research exists in the kombucha drink industry, with a specific focus on drinks quality and users experience rather than pellicle production. In general, little academic research exists on the culture of the pellicle, but for the cultivation of bacterial cellulose in the life science industry (Rajwade *et al*., 2015), food applications (Lin *et al*., 2020), marginally in paper production (Zhang *et al*., 2019), bio-based sensors (Wei *et al*., 2010) or functional aerogels production (Zhen-Yu *et al*., 2014).

Our interest lies in the low cultural anchorage of kombucha pellicle; as there is only a developing set of craft practices developed around the SCOBY, and especially towards drinks production, we are in the presence of an alien material. As no cultural practices are tied to the pellicle yet, it favours its appropriation by anyone, turning it into a peculiar means of experimenting otherness in design.

3.2. Crafting for otherness

thr34d5, a design research NGO, has been developing an inclusive design practice since 2018 through its kombucha Applied Research Program (kARP). Initially focusing on the alteration of the kombucha cultivation process to build an understanding of the pellicle growth, researchers involved in the kARP tested various compositions of the cultivation mix in an attempt to make the process even more sustainable by replacing the sugars provided as nutrients to the SCOBY (glucose and fructose sources). Tea polyphenols are also key to kombucha production, especially as they provide nitrogen to the bacteria. But as tea has an important environmental impact because it is only produced in central and south Asia, our researches also look into replacing it with vernacular organic food wastes as a mean to lower it. Among other areas, research has been conducted on colours, growth containers, scale of production, and waterproofing of the dried pellicle. Experiments are documented on various social media[3] and a git website[4]. The adventurous DIY approach, led by a collaborative will to

[3] Applied research traces can be found on https://instagram.com/thr34d5, https://facebook.com/groups/kombuchafr, and https://facebook.com/thr34d5.

[4] https://wikifactory.com/+thr34d5/karp (consulted November 2019).

build an emergent craft practice of this alien companion, led to the discovery of a variety of material languages, textures and behaviours. While welcoming new researchers in thr34d5' workshop and at workshops animated by thr34d5, the sense of fascination and disgust - to say grossness - that fresh kombucha pellicles trigger is particularly worth noticing. The candid approach to the material in previously mentioned experiments uncovered a wide variety of fascinating textures.

One thing that is learnt from experimenting with biological companions in a spirit of inclusiveness is the respect of its agenda. There is a necessary discussion with the material. Kombucha does not do what we want, it rather agrees to collaborate with us. Kombucha is ours as long as we are theirs. Hence, there is a necessary indeterminacy in the craft practice that is developed. As opposed to a modernist way of "French gardening" the material, liquid design practice aims at leaving space for both the micro-organisms in presence and the social context to express themselves, "English gardening" style. If a slick and eye pleasing design is being curated in galleries for privileged clients, a gross design is activist and its craft aims at giving a voice to the unprivileged, to the marginalized or unheard. Until it is acknowledged that there are no merges, but only situated cultures.

Designing with the oddest, grossest materials, is a political stand in favour of including others, favouring otherness and openness for the sake of not only reconnecting to the territory but doing it collectively.

To sustain and spread the values it promotes, thr34d5 has structured a knowledge transmission sequence throughout workshops on kombucha crafts - growing a pellicle, treating it, making from it. The process itself is particularly simple, with few steps and tolerant of variation - as an example the bacteria need sugar but are quite lenient on the type of sugars provided, which has a surprising influence on colour, texture and thickness of the pellicle. The kARP has been the birthplace for a series of objects made out of kombucha pellicle, from garments to art pieces, all of which contributed to develop novel craft practices. More recently, thr34d5 has started investigating the possibility of harvesting both the pellicle and the kombucha drink to reduce waste from each of the processes. The kARP is an open-source program for participants around the world willing to contribute, and thr34d5 documents the knowledge gathered, making it available online, including the data on kombucha culture and crafting.

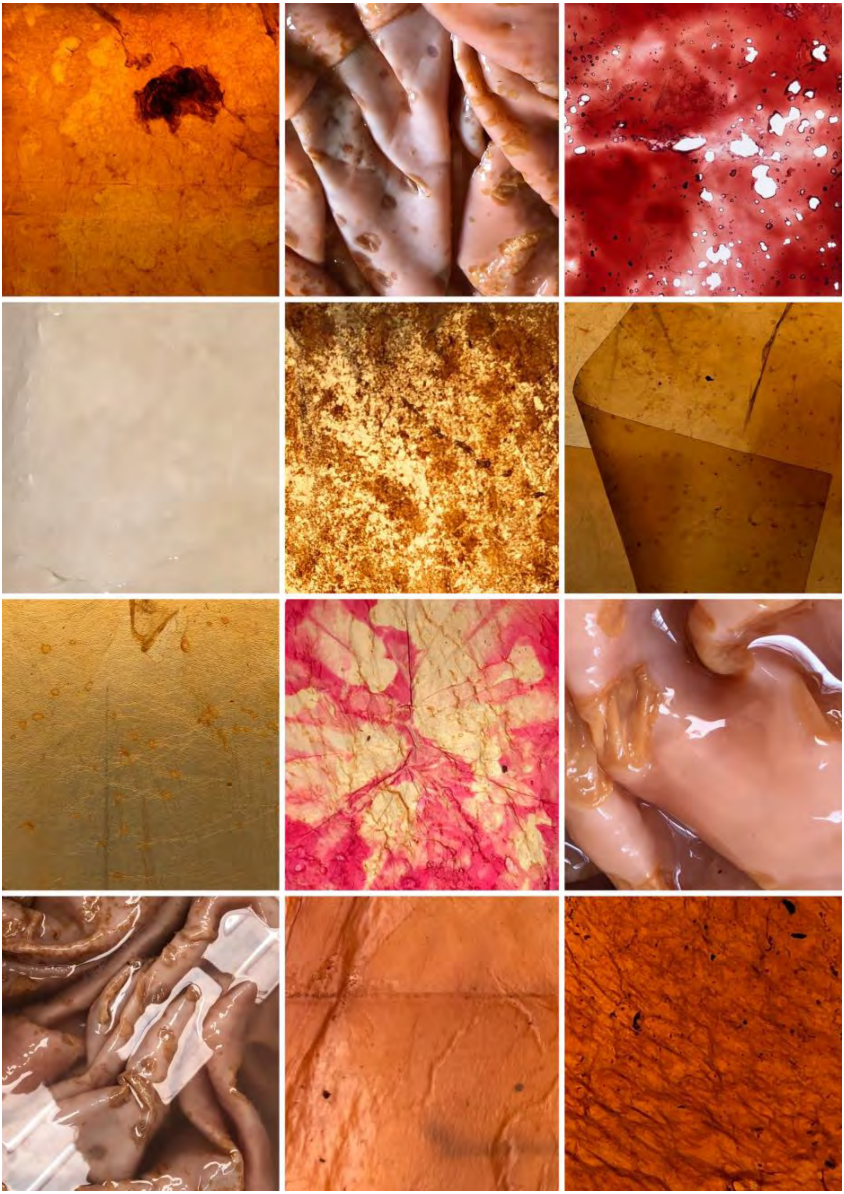

Figure 1. Textures of kombucha as experimented by thr34d5.

4. What is gross is liquid

4.1. Disgust in kombucha

The kombucha pellicle is appalling as much as it is appealing to the five senses, contributing to trigger core disgust (Rozin *et al.*, 2008) (Figures 1 and 2). While the privileged senses of taste and touch are identified as provoking disgust, smell and sight also participate, and the confrontation with the pellicle therefore is a memorable experience. As a pellicle grows in a liquid usually considered a beverage, taste immediately comes into play. A superposition is created with the liveliness of the culture of bacteria when being involved in the process of production; this aspect is deemed particularly gross in Europe where the kombucha drink is mainly sold pasteurized, while much easier to accept in South America for instance where the drink is sold with cultures still active - a reminder that disgust is as much a social construct as is edibility. Whether it is possible to eat the pellicle is also an extremely frequent question, and is in fact possible, with some people even making candy out of it. Thus, the idea of ingestion is constantly present when interacting with the kombucha pellicle. Furthermore, a sweet acidic smell is developed during the long fermentation that is necessary to grow a kombucha pellicle. This characteristic smell is quite familiar to fermented food inclined people as it relates to umami – a flavour found in preparations such as soy sauces, cheeses, wines, pickles, to name a few. The sense of hearing is involved too: while not particularly significant on its own, the squish of the pellicle participates in the general characteristics of an encounter with kombucha cultures.

The pellicle goes from squishy and wet, saturated in water, when retrieved from the mix, to sticky while drying to smooth and soft once treated. The confrontation is constant as the pellicle needs to be manipulated at all stages of the process. Most designs with the pellicle would be garments due to its mechanical behaviour (bacterial cellulose can be up to fifteen times stronger in tensile strength than leather), which involves a skin contact while worn. Skin contact has been shown to be a major component of the gross related emotions. The appearance of the pellicle, reminding skin through its colours and textures, the marks and moulds sometimes appearing during the growth process, the bulges forming when manipulating it wet, are all classic elements of the Grotesque aesthetics.

4.2. From fascination to animism

“When bodies spill out of their boundaries, or when parts are severed from the whole, they become unsettlingly other” (Thackara, 2019). As is shown in the photograph below (Figure 3), a raw kombucha pellicle has a distinct appearance reminiscent of skin and organs. This visual analogy to human guts and body parts leads to a renewed reaction of disgust related to anthropopathism. This proximity to human anatomy can even lead to one not hesitating to confer life to the pellicle. This mechanism reminds of animism and ways to confer a sacred status to non-humans, or life. The friction stemming from the awareness of the possibilities of eating and drinking the resulting liquid and pellicle and of this organic aspect results in the notion of ingesting the other, a feeling of a major transgression of moral boundaries. A path exists on which we could embody the SCOBY, transgress the moral rules to become one.

Figure 2. Endeavor on the aesthetics of fresh kombucha by thr34d5.

The kombucha pellicles, because of their usage as vectors of transmission of SCOBY colonies from peer-to-peer, are also referred to by the terms *mother* and *daughter*. A mother is the strain used to inoculate a kombucha mix and a daughter is the pellicle produced by this mix overtime. Keeping track of the strains and their ties is done in the form of a family tree by thr34d5 as well as by other kombucha researchers, with each mother and daughter baptized with a name and family name. Participants to kARP workshops and to the applied research program are often found to spontaneously baptize their batches of kombucha and refer to the SCOBY as beings. Furthermore, the care for conditions of culture and the growth over time act as a constant reminder that the SCOBY are living organisms. The precautions to keep it from being contaminated during the cultivation setup evoke the fragile aspect of life. Feeding the SCOBY with sugars and witnessing their reactions to changes in the conditions recall that it is not handleable, it is other, and one builds a respect towards it.

4.3. Towards inclusive cosmogonies

While the characteristics of the kombucha production naturally render it gross, thr34d5 has been documenting the kARP activities by further staging the grossness of the process and of the raw material. The lab-like installations where the pellicle is grown, the glass jars in which some are cultivated, also feature a Grant Museum, curiosity cabinet-like style known to be a marker of the Grotesque. Enhancing the flesh and wet aspects of the pellicle stirs fascination, grasps the gaze. The frequent overlap with human bodies seeks to remind that the gross craftsmanship is inclusive by essence, as there is a trans-species collaboration at play. The establishment of a collaborative craft with a non-human is in fact not really new territory, for there are countless works acknowledging the non-continuity of the concept of nature in pastoral ways of living and pre-modern societies (Descola, 2005).

The culture of kombucha pellicle is the place of a lasting clash between disgust and familiarity, generating a sense of fascination and grossness. The personification of the kombucha contrasts with the unsettling solicitation of the five senses during the process. When harvesting and treating the pellicle, it changes from a strange, raw, wet, lively, organic matter to a leatherlike, much more familiar textile. The steps of the process relating to the growth of cellulose, although presenting similarities with producing food and cooking, can be exotic to many given that it involves bacterial culture. The steps relating to the pellicle treatment, however, are commonplace and mobilize everyday tools and activities such as sewing. Kombucha culture therefore goes back and forth from disgusting to usual, in a continuous disparity between liveliness and object-like familiarity. These clashes enhance curiosity towards kombucha, strengthen the impulse to adopt and personify and are an encouragement towards interspecies collaboration. Conversely, kombucha crafts oddly resemble the aesthetics of gut clothing traditionally found in Arctic regions (Schmidt, 2019), which brings an interesting perspective onto our discussion onto aesthetics, taste, craft, transmission and situatedness.

5. Conclusion

The kombucha practices introduced by thr34d5' kARP illustrate a xenodesign practice in that it acknowledges a collaborative dimension of conception in regard to both humans and non-humans by its material semiotics. The program and the introduced design practice focus on care, maintenance, and inclusiveness by adopting an aesthetic of grossness and methodologies fostering indeterminacy and holisticness of senses. Mobilizing the gross in design works is an opportunity for the continuation of the challenging of dominant models, of which the normalized representation of the body (Figure 3). Liquid practices in everyday designs have the potential to carry out activist and affordance aware practices, or rather build impact for designs endorsing intentions. If a user-centric practice is by definition anthropocentric, modernist and replicating dominant models by conforming to what is expected of it, a community-centric practice challenges the establishment, opens the dictionary of semiotics in design (Rigobello & Gaudillière, 2019) and leaves space for identities to become familiar within the relational space that is generated.

Beyond the materiality of kombucha, the present paper explores the potential of a design methodology in becoming building on queer art investigations of identity and otherness, of difference and individuality, and on gross as a reaction, towards the articulation of a design practice for social inclusion, referred to as xenodesign. We understand from the experience of this research that the transmission of knowledge in design and craft practices embedding a such qualities shows much potential for developments, for there is an intuitive and leverageable act of animism happening over the variety of situations encountered that illustrates at individuals' level the striking power lying in material semiotics. This foreseeable aspect of craft practices development should be further investigated with a variety of materials in an effort to build an understanding of knowledge transmission, belonging and intuition for social inclusion.

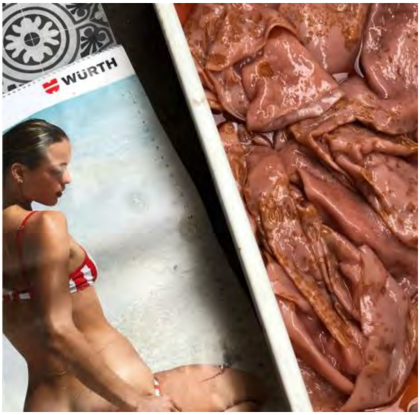

Figure 3. Confrontation of aesthetics by thr34d5.

This research aims at supporting a larger movement in maturing everyday design methodologies to build upon ones that satisfy only a few senses by an “eye-candy” effect or mind pleasing by normalization and identification, that is by encouraging a richer and yet affordable understanding of the material semiotics at hand for contemporary designers.

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