Mycelium Materials

by

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Abstract

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Mycelium materials are made of fungi. By digesting waste to create a bio-composite of mycelium and the feedstock, they are becoming a sustainable material option. Fungi are nature's recyclers, with millions of species that can digest everything from cellulose to polymers. By selecting specific species to digest industrial waste such as sawdust, bio-composites can be grown that can be disposed of after use by composting. Composting reduces their environmental impact in comparison to synthetic composites. As part of an academic-industry collaboration, bio-composites were explored made of *Ganoderma lucidum* fungi. The findings provide researchers and commercial developers with information on the properties, performance and suitable applications of these novel materials. The material was characterized from a physical, mechanical, thermal, and manufacturing standpoint using American Society of Testing and Materials standards. The ultimate tensile strength was 176 kPa, with a tensile modulus of 1.3 MPa, and an ultimate compressive strength of 490 kPa. These properties are comparable to brittle, cellular materials such as expanded polymer foams. The values for the thermal resistance and thermal conductivity ranged from 0.0014 to 0.0019 m^2 K/W and 0.053 to 0.077 W/m K respectively, which are comparable to current natural insulator materials such as balsa wood and natural cork. The average maximum use temperature was found to be 390 ºC (734ºF). These properties are comparable to polymer foams such as expanded polystyrene foam, suiting the material to non-structural applications such as packaging and insulation. Combining a mycelium material core with carbon fiber and bamboo laminate skins improved the flexural strength. The core shear ultimate stress was 36.2 kPa for the mycelium material alone, rising to 63.3 kPa with bamboo skins and 76.6 kPa with carbon fiber skins. The large improvement in strength suggests that with development mycelium materials could be suited to higher-performance applications such as interior wall panels and construction bricks. Methods were tested to manufacture mycelium materials, finding the material could be shaped with drilling, milling, sanding, and that laser cutting can form features of ~3mm. Finally, current and future applications were considered using three use case studies: as packaging, as fabrics, and as a sanitation solution to digest human waste to create useful products. Overall, mycelium materials show great promise for further development as a bio-composite that can digest waste to create useful materials.

Table of Contents

List of Figures

List of Tables

List of Symbols & Abbreviations

ºK	Degrees in Kelvin
$\bar{\rho}$	Density of the mycelium foam material
ρ_s	Density of the mycelium hyphae
GPa	Giga Pascals
g	Gram
Q	Heat flux
kg	Kilogram
kV	Kilovolt
MPa	Mega Pascals
m	Meters
µ	Micro
mm	Millimeters
E_s	Modulus of the mycelium hyphae
$\bar{E}$	Modulus of the mycelium foam material
pA	Pico Ampere
σ	Stress
Th	Surface temperature of hot surface (ºK)
Tc	Surface temperature of cold surface (ºK)
L	Specimen thickness (m)
W	Watts

Introduction

The demand for sustainable materials continues to grow. This increase is due to the increased understanding of how oil-based materials harm the environment, and also the rising pressure from consumers to have access to less harmful materials. In addition, the continued commercial trend towards a circular-economy has as created an opportunity to develop materials which can convert a range of waste into new, useful commodities (Allwood et al., 2001).

New material technologies are being considered by academic and commercial ventures that could mature into alternatives to oil-based materials, including fungi (Bringezu & Bleischwitz, 2017). Fungi-based material technologies have existed for thousands of years, as foods, medicines, and even clothing (Pegler, 2001), but only recently have become a research focus in academic and industry.

This research explores and develops fungi-based technologies from a sustainable materials outlook. It also provides an example of an academic-commercial collaboration between UC Berkeley and Professor Philip G. Ross, co-founder of MycoWorks, a San Francisco based fungi-technology start-up. This research helps create alternatives to polymeric, energy-intensive materials. The range of mycelium-based materials explored in this research are shown in Figure 1.

Figure 1. Various mycelium materials

While naturally occurring fungi materials do not have the mechanical performance needed for high-strength applications, their environmentally low-impact and sustainable characteristics suggest a range non-structural, apparel, and furnishing applications. Tailoring the material to the specific need is also possible due to the material being a composite, making it possible to vary the composition and structure to meet various goals. An example is using the mycelium material as a core for a sandwich composite, combining the light-weight, low-strength properties of the mycelium-based core, with the high-strength properties of bamboo skin laminates. This combination improves the mechanical performance of the mycelium-based material, while also offering less environmental impact during the product lifecycle than oil-based plastics, due to being made of natural materials. Current applications for mycelium materials show a wide range of material properties, applications and relative benefits and costs in comparison to similar natural & synthetic materials, as summarized below in Table 1.

Material	**Modulus, E (MPa)**	**Density, ρ (kg/m³)**	**Ultimate Strength, S_{utc} (MPa)**	**Supply avail-ability**	**Cost**	**Environ mental impact**	**Applications**
Present research *Ganoderma lucidum composite*	1.30	318	0.49	Low	$	Low	Apparel Low-strength sandwich panels Furnishings Packaging
Starch-based foam (w/ fiber)	183	260	1.09	Mediu m	$$	Low	Furnishings Packaging
Polystyrene foam	5.70	41.2	0.18	High	$	High	Automotive Furnishings Packaging
Polyvinyl-chloride foam	3.00	500	45.0	High	$	High	Apparel Automotive Furnishings
Aluminum foam	347	255	1.24	Mediu m	$$$	Medium	Aerospace Automotive

Table 1. Mycelium materials & comparative characteristics
Summarized from Table 2 and (Stevens et al., 2010)

Comparatively, the stiffness & strength of the mycelium material made from *Ganoderma lucidum* fungi are orders of magnitude away from a high-performance foam, such as shown in Table 1. This shows the material is not ideal for high mechanical performance applications such as structural insulated panels for homebuilding, but also shows it could be used as an alternatives for many of the lower mechanical performance applications that synthetic foams are currently used for, such as packaging and insulation.

The majority of current research on mycelium composites is for commercial purposes. Thus, the information remains as trade secrets. This has not facilitated the sharing of information and growth within the field, and the disparate nature of the research efforts, ranging from collaborative efforts to hobbyist projects, would benefit from a centralized network of knowledge. This research aims to provide a curated set of research topics focused on mycelium-based materials, to explore the fabrication, manufacture, mechanical & thermal performance, and current and potential commercial applications of this novel material. To understand the performance, design capabilities, and potential applications of mycelium materials, the physical, mechanical and thermal properties of a fungi-based composite material were experimentally determined, based on American Society of Testing and Materials (ASTM) testing methodologies. Many novel fabrication methods were developed during this research process. A secondary aim of this report is to help facilitate fellow researchers and developers by sharing these novel methods and results.

The hypothesis is that mycelium materials can demonstrate properties to be a suitable replacement material for applications that currently use polymeric materials. Relevant properties include

- Mechanical & thermal properties
- Manufacturing & fabrication properties

Given the properties I identify, my third section discusses several possible applications. Each of these three areas are explored in three main sections of this research report, ending with a conclusion that summarizes the key findings, and considers the potential of mycelium material for future research.

Chapter Overviews

Chapter One As mycelium materials are biological in nature, the importance of being a natural material will also be considered, including discussing the composition as a fungi-based material, and the wider background of fungi and their properties.

Chapter Two explores the research completed to understand the structure and composition of the cellular material. This included using optical and scanning electron microscopy to identify its cellular structure, and the use of a model to predict cellular properties.

Chapter Three explores the mechanical properties of mycelium materials, as a key component of the research efforts to support the development of fungi-based sustainable materials, by considering the material's properties and resulting design capabilities, and comparing the performance to similar natural and synthetic materials.

Chapter Four determines the thermal properties of the mycelium material through testing, helping create information for designers on the maximum use temperature and materials performance as an insulator, as the cellular structure properties lend them towards insulation applications.

Chapter Five focuses on the flexural performance that was explored through the testing of sandwich composites with a mycelium material core, with the goal of producing higher performance properties. While entirely natural materials allow for greater sustainability and lower-impact manufacturing and disposal options, both synthetic and natural materials are considered to fully explore possibilities.

Chapter Six addresses the mechanical manufacturing of mycelium materials, as these properties and capabilities are equally important to researchers and designers. Information on suitable methods to shape the material will inform the creation of testing samples and products made from mycelium materials.

Chapter Seven explores thermal cutting methods as an alternative to mechanical cutting methods, using experimental methods. The performance of thermal based cutting methods in fabricating small features was evaluated, including the suitability to create small features, which is a limitation of mechanical cutting due to the frangible nature of the mycelium material.

Chapter Eight considers the degradation of the material in interior and exterior environments, inform designers and developers of the material durability, and considers options to increase it.

Chapter Nine discusses the organic growth of the mycelium material as a biological material including its biological growth parameters, denaturing methods required, and a brief discussion of the characteristics of fungi that impact their growth as a manufactured object.

Chapter Ten considers current and developing applications of mycelium materials. Two current efforts to commercialize mycelium materials for consumer applications are introduced in the form of two case studies; packaging and natural biomimicary of leather.

Chapter Eleven explores the future applications of mycelium materials, through a case study of the potential for mycelium materials to be used as a sanitation technology in low-resource countries. Fungi's natural aptitude to breaking down and recycling a huge variety of organic and synthetic materials lends them towards challenges where conventional sanitation technologies are unsuitable.

Finally the conclusions and further work summarizes the key findings of this research effort. Having determined overall that mycelium materials have suitable properties to replace natural and synthetic materials in non-structural applications, they show good potential for further development and further avenues of investigation are suggested.

SECTION ONE: FUNGI MATERIALS

Chapter One – Fungi as a an Emerging Technology

Fungi, Mycelium & Their Potential for Sustainable Materials

The environmental impact of synthetic materials continues to be a growing topic of concern to both manufacturers and consumers, and novel bio-composites are being developed to provide alternative options (Mohanty et al., 2002). The development of bio-based materials continues to be driven by the need to reduce the environmental impact on our planet. The development of materials made from fungi, known as mycelium composites, is becoming an emergent field in the range of bio-based materials being developed. Motivated by commercial and consumer demand for less ecologically damaging options, mycelium composites continue to grow as a sustainable materials option, offering reduced environmental impact due to being made from carbon-containing waste by-products from industrial processes. Over 50 million tons of cellulosic and organic waste is available for recycling annually in the USA (United States Environmental Protection Agency, 2014) in the format of cardboard, wood matter and organic compost matter. All of these carbon containing materials are suitable feedstocks for fungi, which can digest them and use them as nutrients through enzymatic interactions (Eriksson et al., 1974). The use of mycelium materials in artisanal and commercial applications continues to increase, including plans for use of the material by one of the world's largest furniture retailers, IKEA (Gosden, 2016), leading to further demand for information on the characteristics of mycelium materials.

Fungi have their own classification kingdom, due to having traits of both plants and animals. There are millions of fungi species (Blackwell, 2011) each specializing in consuming a particular type of matter. This can range from organic matter such as wood, to synthetic materials including polymers and fiber reinforced composites (Barratt et al., 2003), (Cosgrove et al, 2007) and (Gu et al., 1997). Fungi have a natural ability to digest carbon-containing materials and transform them into structures made of chitin (Hammel, 1997) a natural fibrous polymer (Rinaudo, 2006), through the formation of a network of mycelium hyphae (Carlile, 1995), (Finlay & Read, 1986). The hyphae are made of chitin, which is found in crustaceans, coleopteran (beetles), and the cell walls of fungi. Chitin can take on soft, flexible forms such as in the joins of arthropods, or hard, rigid forms such as the exoskeleton of a beetle (Zhang et al., 2000). This versatility lends itself to the creation of a natural biocomposite, with the composite matrix being formed by the mycelium, and the composite reinforcement being formed of the original feedstock. Volume fractions of the matrix and reinforcement can vary significantly with the selected degree of growth of the material, and composition of the reinforcement.

Mycelium materials are formed of the vegetative part of fungi, mycelium, which is a eukaryotic cell encapsulated by a rigid cell wall. This forms the filamentous cellular structure (Gibson and Ashby, 1999) through biological growth, which gives the materials their ability to grow into a molded shape. The material is a bio-composite of the fungi organic matter and the remaining undigested feedstock the fungi fed on. Various feedstocks can be used to grow the myology materials, including waste agricultural matter such as lignin-based sawdust (Hammel, 1997), chaff, and other natural byproducts. The feedstocks are combined with the selected species and placed in a growth environment for several weeks, during which the fungi species digests the feedstock and forms a cellular structure through the growth of mycelium. Mycelium is the vegetative part of a fungus, and is a mass of branching, thread-like individual hyphae. The growing unit of the mycelium material are hyphae, which provide the growth, locomotion, nutrient-seeking, digestive, reproductive and communication functions of the fungi. The mycelium hyphae network is used to grow to move location and seek new feedstocks, uptake and transport nutrients, and to relay chemical signaling mechanisms which govern the mycelium architecture, growth and all other functions (Finlay et al., 1986). Hyphae are remarkably multifunctional, even including hyphae used as a predatory tool. Some fungi species predating on animal species, such as the predatory *Orbilia* species of fungi which prey on nematodes (Malloch, 2017), (a type of worm from the phylum *Nematoda*, such Roundworm), using hyphae that form 'lasso' loops in which the nematodes become entangled in, and are then digested (Barron, 2003). Although not explored in this research, fungi sometimes harm humans and other animals. Examples are the parasitic relationship with plants (such as fungi that have specialized hyphae that penetrate and remove nutrition from the host plant (Mendgen & Hahn, 2002), and parasitic relationships with animals (such as disease causing fungal infections in both humans and animals). Hyphae branch to fill the space available (Edelstein & Segel, 1983), as long, branching filamentous structures, and grow throughout the material and form a large fiber network (Bulawa, 1993). Hyphae grow into a cellular material from the inoculation of a mycelium fungal strain spore, consuming carbon and nitrogen containing feedstocks to build a network of mycelium hyphae (Carlile, 1995). Growth and nutrient harvesting are achieved by the secretion of digestive enzymes, dismantling macro-molecules into components then absorbed using a diffusion gradient or by transport mechanisms. Unlike plant matter based on cellulose and containing rigid lignin, rigidity in fungi is achieved with chitin composed of polysaccharides, which forms the skeletal structural of the fungi and defines the mycelium cellular structure. The growth can be controlled through regulating the growing environment, including the physical space, and the biological surroundings such as intensity and availability of light, water and gaseous exchanges (Chau, 2016). Mycelium has a fast growth rate and will continue to grow without limitation with sufficient nutrients - the largest known mycelium growth is in eastern Oregon, USA, covering over 2,400 acres and estimated at over 2,200 years old (Stamets, 2005).

Unusually many fungi species are non-photosynthetic organisms that absorb all their required nutrients from their host feedstock and environment. Typically found buried within degrading and rotting bio-matter including wood, leaf matter, flesh, or any other carbon-containing matter, the mycelium structure of fungi is often hidden within the host matter and only becomes visible when the fruiting stage is reached and fruiting bodies appear as mushrooms on the surface to disperse spores. In this investigation only pre-fruiting stage mycelium was used for safety, to prevent exposure to spores. Their unique ability to grow in a wide range of environments, while needing specific feedstocks has attracted the interest of materials engineers, seeking replacements for highly processed, environmentally damaging synthetic materials. Using fungi to grow the mycelial form and structure enables the potential for a designed physical geometry for an application, such as an insulation material that can be naturally manufactured (Mayoral, 2011). Thus mycelium materials offer great potential for development into commercial products. Fungi have a relatively fast growth rate and given sufficient nutrients will grow to vast quantities (Stamets, 2005), making them an ideal choice for high-volume degradation and fabrication of materials. Fungi are already commercialized for use as biological control agents (Shah & Pell, 2003) and environmental remediation including the safe capture of heavy metals (Galli et al., 1994), (Baik et al., 2002).

Mycelium are an important part of our ecosystem, and play a vital role in the recycling of minerals and carbon and in the nitrogen-fixing cycle. Edible mycelium fungi are farmed for human consumption, and are used in a broad range of applications including environmental control with fungal biological control agents (Shah & Pell, 2003), capture and safe removal of heavy metal contaminants (Galli et al., 1994), and even medical uses fighting cancer (Wu et al., 2007). As a biological material, mycelium presents an attractive sustainable option replace synthetic packaging and as structural materials with the addition of reinforcements. Synthetic composite materials remain the material of choice for a wide variety of applications, but the high cost of raw materials and complex processing is providing an opportunity to develop more sustainable composites. Current crude oil-based polymeric materials are not sustainable and have damaging environmental impacts including non-renewable resource consumption, toxic byproducts, and the resulting solid waste which is largely disposed of in landfill, storing up problems for future generations (Scott, 1999). For example the natural degradation of polystyrene foam is very slow (less than one percent per year) and results in chlorinated hydrocarbon and other byproducts toxic to animals and aquatic life. Mycelium materials are compostable, and thus could provide a replacement option. An advantage is that mycelium materials are created from feedstocks of agricultural waste such as sawdust and husks, thus providing a sustainable material, and one that reduces the environmental impact of its manufacturing. This also improves the financial implications of developing and selling a new mycelium natural material, as since the material is grown from waste the feedstock costs are reduced, unlike synthetic materials. Due to their high growth rates, non-toxicity and sustainability mycelium-based materials are being explored for use in consumer and artistic applications, including using mycelium-based building blocks for avant-garde structures termed Mycotechure (Ross, 2013), shown in Figure 1. Mycelium materials have also been proposed as a living architecture material, for a prototype sustainable building for recreation in Kunming, Southern China (Phiriyaphongsak, 2013). Several applications for fungi-based materials are already commercially available. West-coast based MycoWorks, who also collaborated on this research, and east-coast based Ecovative Design LLC, supply packaging and textile products designed as alternatives to expanded polystyrene packaging and leather respectively. With the packaging market worth an estimated $2 billion dollars (Bliss, 2013), bio-based alternatives are gathering momentum as they offer advantages of compostability and reduced impact.

Figure 2. 'Mycotectural Alpha' by Professor Philip G Ross, formed of mycelium-based blocks, exhibited at Kunsthalle Düsseldorf as part of the Eat Art Exhibit 2009 (MycoWorks, 2016)

Chapter Two – Characterizing a Cellular Mycelium Material

The novelty of mycelium-based materials means there is an unmet need for empirically determined material properties, as currently few points of information have been created. This in turn requires an understanding of the structure and resulting physical properties of the mycelium-based materials, which have a cellular structure. This research identified key material properties such as density, and explored the structure using optical & scanning electron microscopy, and based on these results, developed of a model to explain the deformation of the cellular structure under load, and predict the material's Youngs Modulus.

Exploring Mycelium Growth Variation with Optical Microscopy

Several images were taken using a scanning electron microscope (SEM) and an optical microscope, to determine if there was variation in cell structure or growth direction. Optical microscopy was carried out on samples from the longitudinal and transverse directions of the mycelium block, at 4X and 8X magnification. Specimens of 25.4 mm (1.0 inch) in cubic shape were prepared for optical microscopy, from sections transverse and longitudinal to the direction of growth of the mycelium material, which correspond to across and with the overall direction the block of material grew. Two samples were cold-mounted in resin and polished for optical microscopy, and two samples were left unmounted. These images enabled the examination of differences in the wood-mycelium composition throughout the sample, and also the examination of the cellular structure of the mycelium and the wood separately. The optical microscopy images, seen in Figure 3, were taken in the transverse (a) & (c) and longitudinal (b) & (d) directions of the blocks of mycelium material.

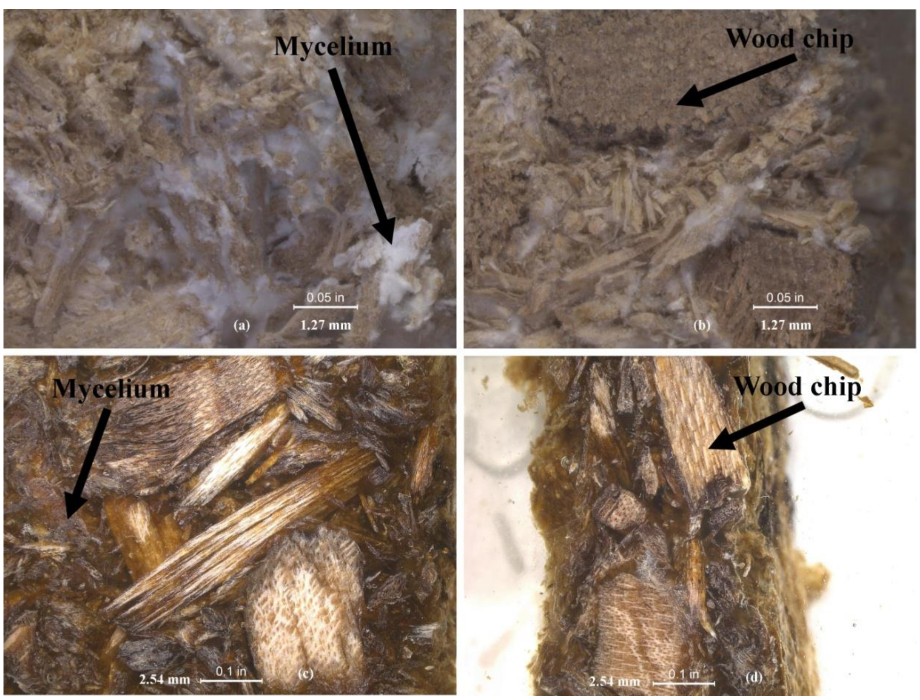

Figure 3. Resin-mounted optical microscopy images
(a) Unmounted transverse image at 4X (b) Unmounted longitudinal image at 4X
(c) Resin mounted transverse image 8X (d) Resin mounted longitudinal image at 8X

The images in Figure 3 show clusters of undigested wood feedstock with clearly identifiable longitudinal wood cells. The resin-infused microscopy samples were found to better highlight the feedstock wood chips, but obscure the mycelium growth areas. This is likely due to the resin infiltrating the wood cells, which are then exposed during the polishing process. The mycelium seen in the non-resin infused samples could be seen to be comprised of many small mycelial fibers, which may be too small to note at 4X and 8X magnification when coated in resin. Image (c) of an un-mounted specimen at the same location better shows the white fibrous interior mycelial growth around the wood feedstock. Neither the transverse or longitudinal samples showed any clearly defined growth orientation of the mycelium material. This is consistent with the previously identified cellular structure of mycelium as non-directional tetrahedrally-connected struts (Ashby, 2005).

Scanning Electron Microscopy of Mycelium's Building Block; Hyphae

Sample imaging was completed using an SEM. Five samples were taken from different locations in a block of the mycelium material, and multiple images at a range of magnifications were captured for each. A Leo 430 SEM was used at a setting of 5.0 kV and around 30.0 pA. The samples were coated with gold-palladium to prevent the material charging and preventing the electron-based SEM imaging. The range of samples were selected to provide information on the format, orientation (if any) and degree of mycelial growth. Samples were cut from a single mycelium-wood block, from five different locations: the center (sample 1), the top surface (sample 2), the bottom surface (sample 5), a cluster of pure mycelium from just under the surface (sample 3), and of an individual wood chip (sample 4), as shown in Figure 4. The images show extreme heterogeneity at all scales, with a randomly oriented mix of mycelium and woodchips. The block of mycelium materials the samples were taken from showed a fully white mycelium-coated surface, resulting in samples 2 and 5 comprising of mostly mycelium. The density of mycelium decreased towards the center of the block the samples were taken from, resulting in sample 4 being entirely comprised of a wood chip. The section just under the block surface was selected for sample 1 and 3, as the mycelial growth was the thickest around the wood chips.

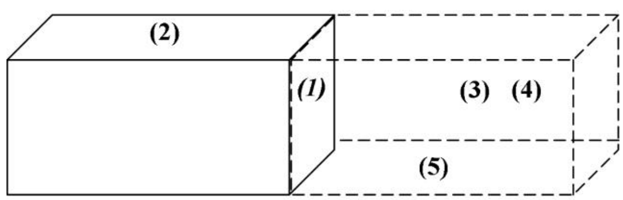

Figure 4. Locations of material sampling from block for SEM images
(1) Interior mixed (2) Top surface (3) Interior pure mycelium (4) Interior pure wood chip (5) Bottom surface

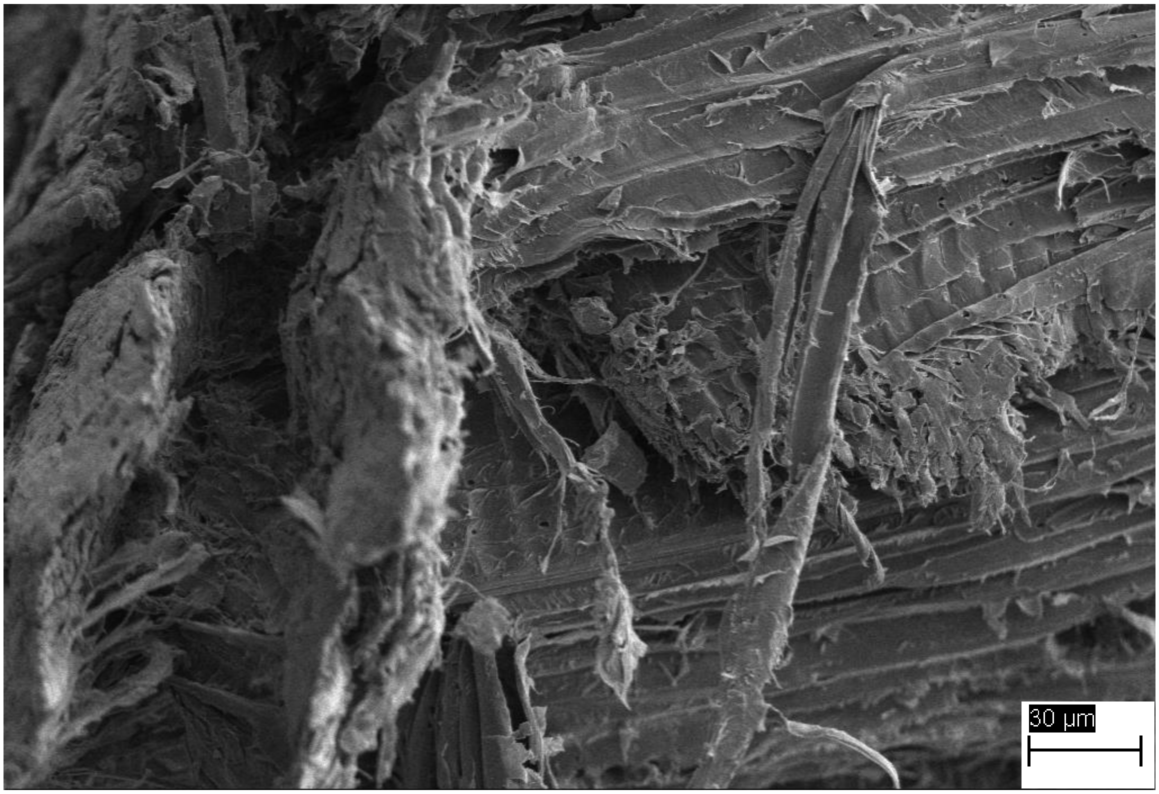

Figure 5. SEM image of mycelium materials sample 4 (interior pure wood chip) X750 magnification

The SEM image of the wood chip, sample 4, showed the well-established cellular structure of wood. Despite the chip being from the interior of the sampling block, surrounded by mycelial growth, a surprising result was the lack of the presence of mycelium hyphae on the surface or visible interior parts of the wood chip. No hyphae were visible in the image, suggesting digestion processes of the fungi species had not begun on the wood chip show in Figure 5. This may explain the frangible nature of mycelium materials, which show brittle fracture between pieces of the material and original wood chip feedstock, as until digestion processes begin there is little to no ingress of the hyphae needed to bond the material together.

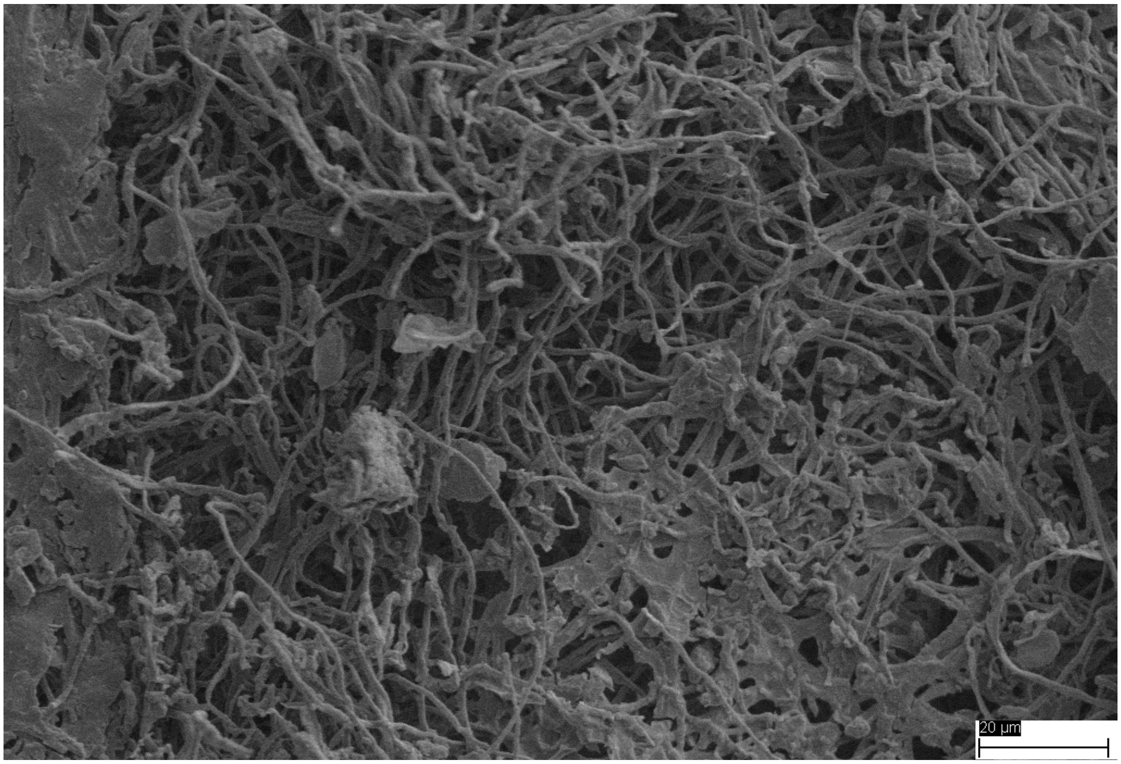

Figure 6. SEM image of mycelium materials sample 5 (bottom) X2040 magnification

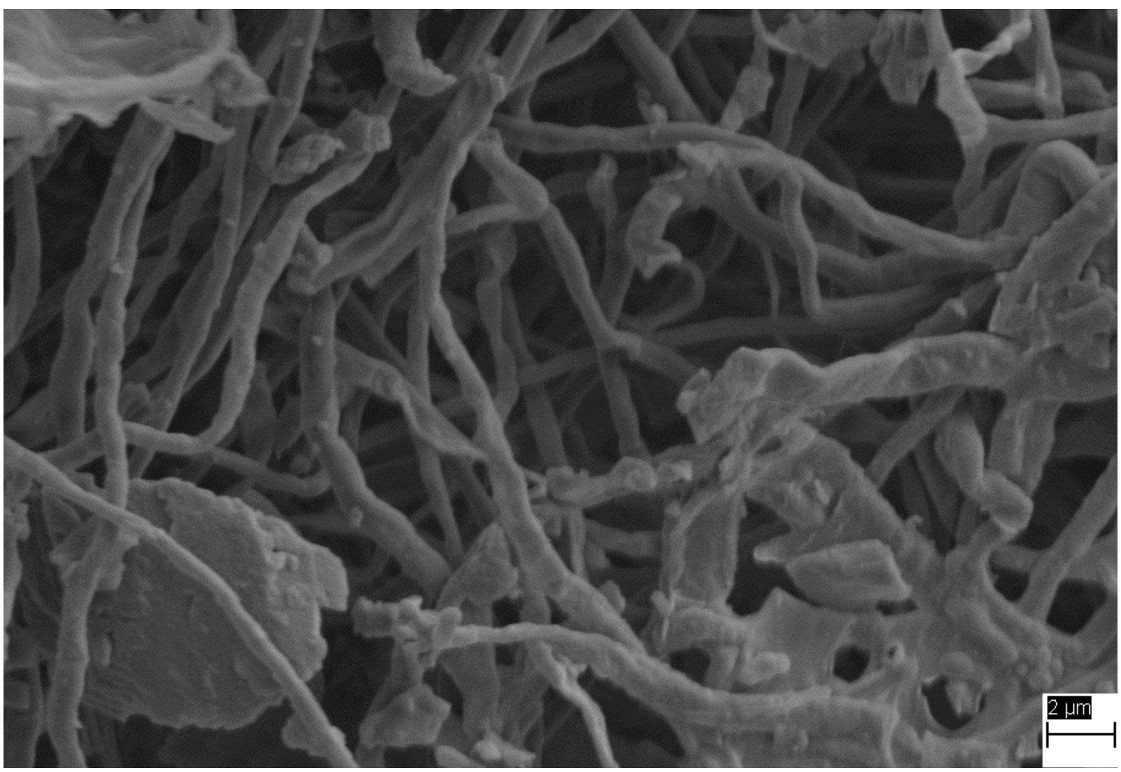

Figure 7. SEM image of mycelium materials sample 5 (bottom) X8070 magnification

The individual hyphae form a tangled, random network, as seen in Figure 6, with both short and long hyphae growing in a variety of directions and crossing over each other and in some cases, conjoining. The structure of the hyphae resembles synthetic random-fiber materials, such as non-woven glass fiber textiles. This may suggest a potential application of the mycelium materials, replacing synthetic fibers with mycelial fibers, which could be reinforced with other natural, biodegradable fibers (such as flax or cotton) – the motivation for such materials is creating a circular economy, where materials are created, used, and reformed, aimed to reduce the ecological impacts of the manufacturing and disposal phase. The image in Figure 6 shows the two distinct forms, of the individual hyphae and of the formation of a continuous layer, in the process of forming the external surface of the sample block as the sample was located on the bottom exterior of the block. To determine if the continuous surface was formed from mycelium, the same sample was investigated at X8070 magnification, shown in Figure 7. It shows (seen in the lower right corner) that the hyphae that are closest to the surface of the sample block thicken and spread, forming a thin sheet of mycelium, that extends between hyphae, as seen in Figure 7. This is theorized to be around the same thickness as the hyphae, and appears to be formed of the same material, chitin – this could be confirmed by comparing interior and exterior samples of the mycelium in a mass-spectrometer, analyzing for differences in the chemical composition. If none was found, it would indicate the mycelium uses the same growth mechanisms, but formed in different ways to produce different material morphologies.

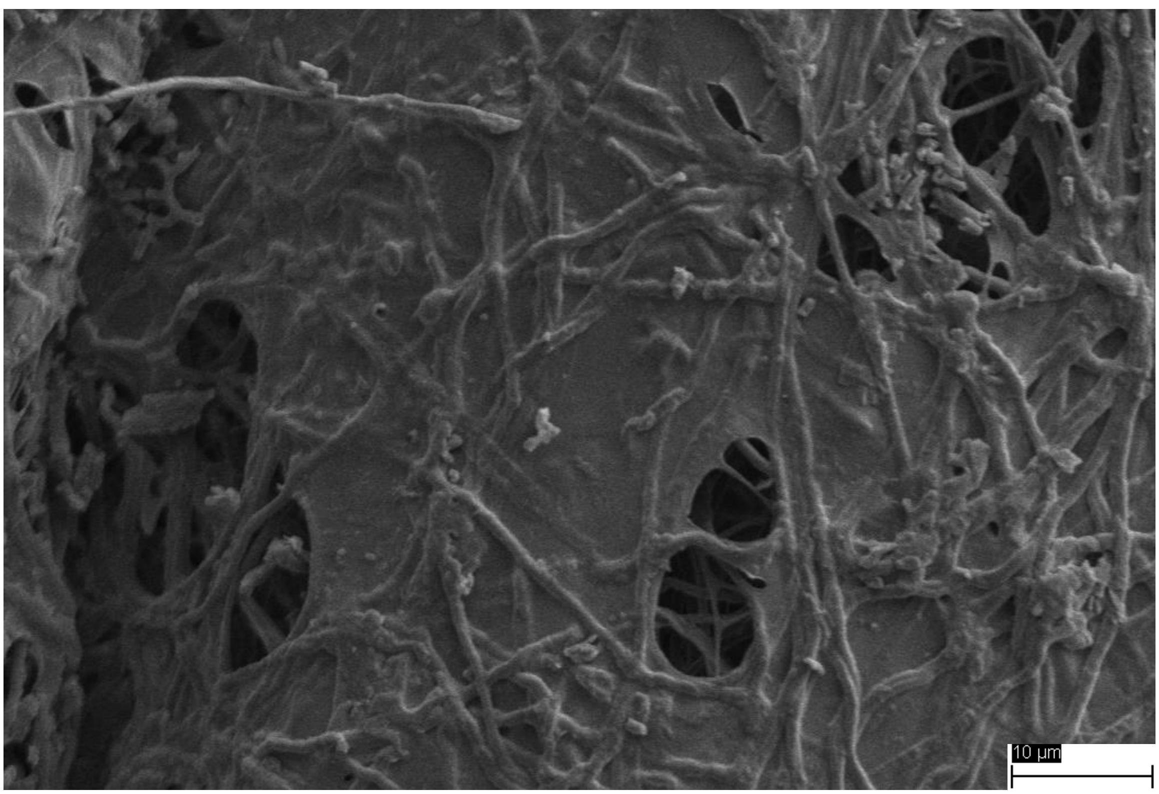

Figure 8. SEM image of mycelium materials sample 2 (top surface) X 3770 magnification, showing hyphae merged to form a continuous surface made from mycelium

The formation of the continuous layer on the exterior of the sample, seen clearest in Figure 8, was only found on the exterior samples, confirming the anecdotal evidence that mycelium materials in molds form a continuous layer in response to contact with the mold surface. This is a natural response of the fungi to meeting an impermeable barrier, through which growth or nutrient acquisition is impossible, potentially to prevent moisture loss. Since the continuous sheet appears to be formed between growing hyphae, it is expected to be the same thickness as the hyphae. Further study into the growth mechanisms of hyphae from a biological perspective would provide more information on the decision-making process of fungi around growth and the resulting shape and form the fungi takes. Since this is a highly complex topic, and a research effort in its own right, this is beyond the scope of this research, but could provide future researchers potential new ways to control the growth and morphology of mycelium materials.

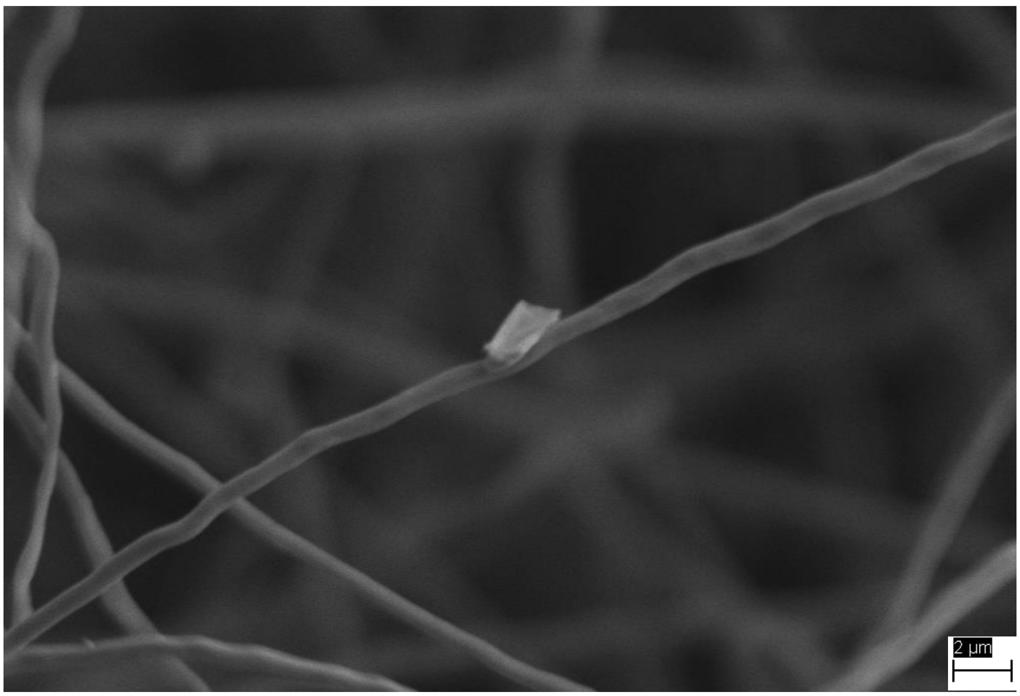

Figure 9. SEM image of mycelium materials sample 1 (mixed interior) X 7906 magnification, showing an individual hyphae growing through the mycelium material.

To determine the thickness of an individual hyphae, a single growing hyphae was identified that had been growing within the interior of the mycelium material, seen in Figure 9 as a denatured hyphal filament, which was originally growing in an aqueous biological fluid mostly comprised of water. The image shows the hyphae suspended in an air space, due to the material being desiccated after the material growth phase. The diameter of the hyphae was calculated from the images, including Figure 9, showing the hyphae diameter to be within the range of 0.5-1.0µm. This is around the range of other reported diameters of hyphae from 2.0-3.0µm (Bhosle et al., 2010), (Li et al., 2015).

Physical Properties

The composition of the material is a mix of the mycelium and the remaining feedstock of undigested wood chips. The material is formed as mycelium digest and form new organic matter.

The network of hyphae penetrates the feedstock's cells, using secretions of acids and enzymes, and converts the nutrients to mycelium, growing the network of hyphae. Areas of pure mycelium growth (white) can be seen within the material, and the surrounding external surface is composed of a continuous layer of mycelium. An interesting observation was that the cellular nature also usefully lends itself to limiting the impact of damage, preventing the structural failure at one location, from impacting the properties and performance at another location, adding to the toughness of the material. Currently there is no established method to determine the ratio of fibrous mycelium matter to wood chip feedstock matter in mycelium composites. Several methods to estimate the relative composition fraction of wood and mycelium were considered, including estimation by analysis of SEM image or visual inspection, but neither method was developed further or tested for accuracy. Analogous to the fiber volume fraction of fiber reinforced polymeric composites, the density of the mycelium material was calculated as an average. An improvement to this method could be considering it as a composite material, with the remaining wood chip as clusters of fiber reinforcements and the grown mycelium as the matrix, so an estimation of the composite density could be made using the rule of mixtures. Since the mass fractions of fiber and matrix cannot be found from the raw materials (due to the conversion of the carbon-containing cellulose into carbon-containing cellular structures), the composite density would only be calculated by estimating the wood fiber and mycelium matrix volume fractions.

Empirically Determined Density

The density of the dried mycelium is known as 1400 kg/m^3 (Van Suijdam et al., 1982), however, experimental measurement, and using the method of dividing mass by volume identified the density as 318 kg/m^3. The difference is accounted for by the degree of digestion of the feedstock into mycelium – the former value is for pure mycelium while the latter is for mycelium with remaining undigested feedstock, resulting in lower density.

Mycelium Materials Cellular Structure

The mycelium materials can be conceptualized as a composite material composed of a mycelium foam matrix, with reinforcing short wood fibers from the feedstock. Wood itself is a highly anisotropic closed-cell foam, thus the composite's hierarchical nature. Based on previous micrographs of mycelium, *Ganoderma lucidum*-based materials were expected to be an open-cell foam (Ecovative Design LLC, 2019a). Previous studies of the mechanical behavior of open-cell foams can be used to inform predictions of the behavior of mycelium. A foam is a three dimensional array of cells forming a cellular solid, such as cork, polyurethane foam, or even bread. Open-cell specifies that the cells have no cell walls, only beams forming cell edges. Liquids and gases can pass more freely through an open-cell foam than a closed-cell, as is required for gas exchange within biological organisms (Gibson and Ashby, 1999). Foams behave differently in tension and compression, and the stress-strain curve also varies depending on the solid material. Figure 10 (Gibson & Ashby, 1999) shows typical curves for compression and tension for three cellular material types: elastomeric, elastic-plastic, and elastic-brittle.

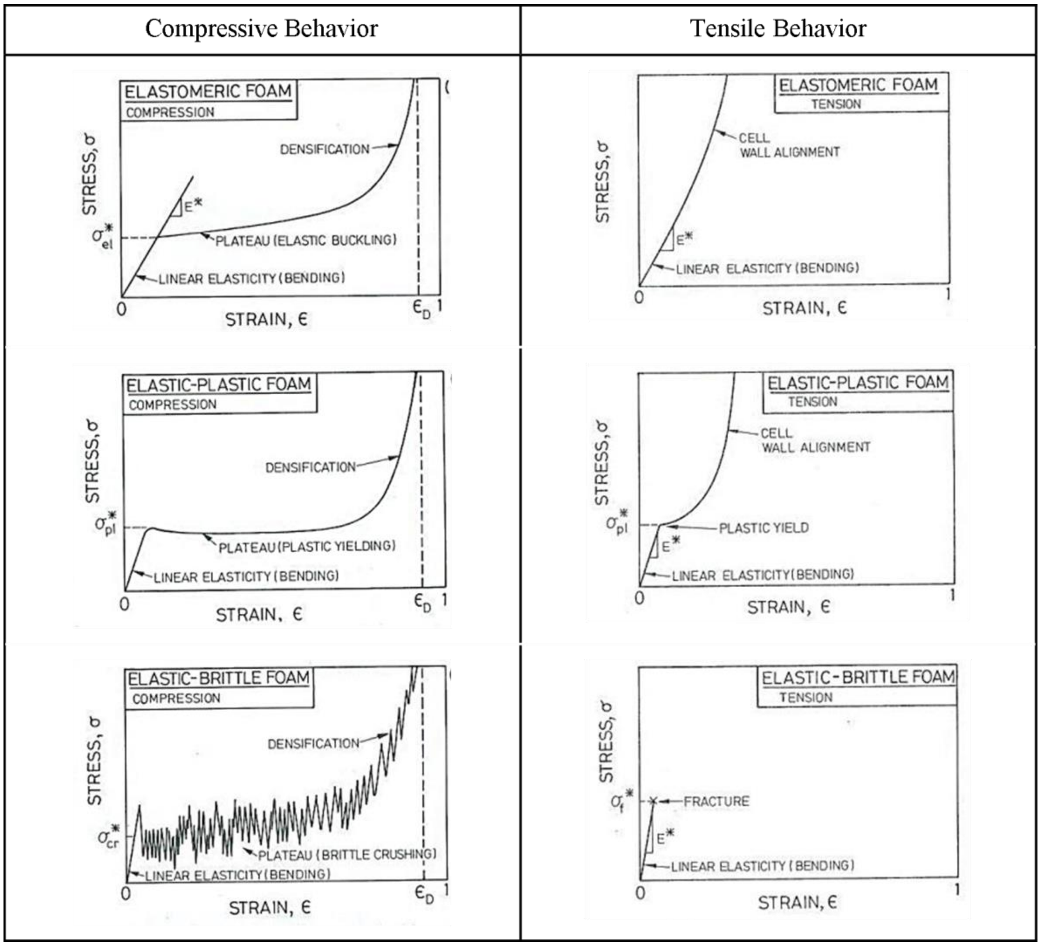

Figure 10. Typical stress-strain behavior in foams (Adapted from Gibson & Ashby, 1999)

The compression curves all show three distinct regions: linear elastic, plateau, and densification. The linear-elastic region reflects elastic bending of the cell edges and stretching of cell walls (if present). The plateau occurs as the cells begin to collapse, via either elastic buckling (for elastomers), plastic yielding (for elastic-plastic materials), or crushing (for brittle materials). Finally, once the cells have fully collapsed, the solid material is compressed and the stiffness increases to fracture during the densification region (Gibson & Ashby, 1999). Tensile stress-strain behavior also begins with a linear-elastic region. For elastomeric and elastic-plastic materials, the stiffness increases as the cell walls become aligned. For elastic-plastic materials, the cell walls must yield first, reflected in a brief plateau. In brittle foams, fracture occurs before the cell walls can align. Some foams demonstrate brittle behavior in tension, but elastic-plastic behavior in compression (Gibson & Ashby, 1999). Deformation of the cubic strut structure occurs under load as modeled in Figure 11.

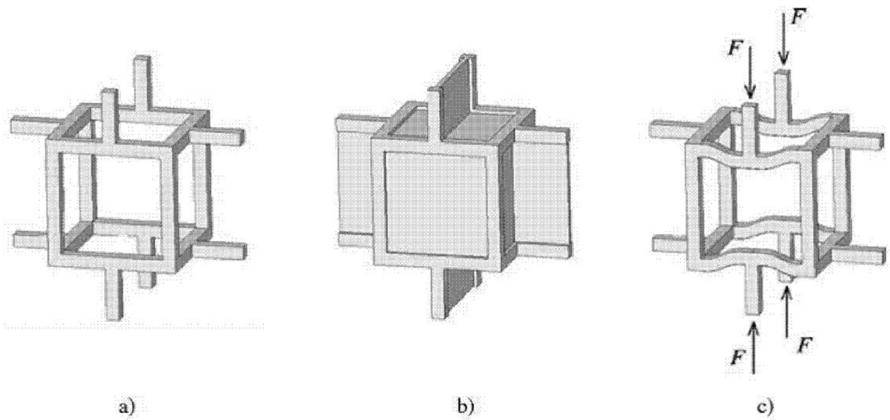

Figure 11. Unit cell: (a) for an open-cell foam of cubic symmetry, (b) for a closed-cell foam of cubic symmetry, (c) shown after linear-elastic deflection (open-cell) (Gibson & Ashby, 1982)

In nature, grown structures of mycelium are not as regular as the theoretical structure, with natural variations of the material due its own biological nature, variations from adaptation to its environment, and natural variation from the feedstocks composition. While the mycelium material is considered open-celled for gaseous exchange, it is biological in nature and may have non-uniform areas of closed-cell architecture, suggesting local geometry of the cell walls may affect the mechanical performance. However, previous research on other materials has shown that most open or closed cell foams act as if open-celled, due to the negligible contribution to the overall stiffness and strength from the thin cell wall faces (Han et al., 1998). Ashby, widely published in describing the mechanical behavior of bending dominated cellular structures, together with Gibson proposed Equation (1) to describe the properties of air filled foams based on the elastic modulus at a specified density (Ashby, 2005), where $\bar{E}$ is modulus of the mycelium foam material, E_s is the modulus of the mycelium hyphea, $\bar{\rho}$ is the density of the mycelium foam material and ρ_s is the density of the mycelium hyphea.

$$\bar{E}/E_s \propto (\bar{\rho} / \rho_s)^2 \quad (1)$$

In order to determine the mechanical properties of mycelium materials from a mechanics of materials viewpoint, the asymptotic homogenization method was selected as most suitable, to understand the materials behavior as a heterogeneous composite. It was chosen as when the microstructure is subject to loads, which vary in comparison to the macrostructure, it results in different effects to the response to loading at different length scales. A length scale is a particular length defined within one order of magnitude – for mycelium materials, these are the micro and macro structures. Mycelium materials also exhibit hierarchical formations, with material properties, structures and morphologies expressed at different scales within the material. For example, in the micro scale a unit cell, modeled as comprising of a single open-walled cube with struts made of chitin, can have different material properties and structures to the macro scale of a cellular material formed of repeating unit cells.

Asymptotic expansion theory, as posed by (Bensoussan & Papanicolaou (2011), and Fish (2009) assumes a length scale which relates both the microscopic and macroscopic material behaviors, simultaneously, using ‘fast’ and ‘slow’ variables. The relationship of the fast and slow variables is shown below in Equation 2, where ϵ is a very small number:

$$\epsilon = \frac{x_i}{y_i} \ll 1 \tag{2}$$

Asymptotic homogenization is designed for periodic structures containing different scales, such as the repeating cellular structure of mycelium materials, which exhibit different properties at different scales. As a composite, the mycelium material has a repeating cubic cell unit structure with two different scales, micro and macro. Thus the asymptotic expansion of a defined displacement field is shown in Equation 3 below:

$$u_i^{\epsilon}\,(x) = u_i^{(0)}(x, y) + \epsilon u_i^{(1)}(x, y) + \,\ldots \tag{3}$$

The expression can have infinite terms for multiple scales, but as mycelium materials have two distinct scales within a unit cell, the equation can be shortened to retain only the first two terms while preserving generality. The equilibrium equation, seen in Equation 4 can be expanded in different scales (Li & Wang, 2008),

$$\frac{\partial \sigma_{ij}^{\epsilon}}{\partial x_i} + \,b_i = 0 \tag{4}$$

And since the Cauchy stress tensor can be defined by Hooke’s Law, Equation 5 is shown:

$$\sigma_{ij}^{\epsilon} = \,C_{ijkl} e_{kl}^{\epsilon} \tag{5}$$

And since C_{ijkl} is the fourth order elastic tensor, and e_{kl}^{ϵ}can be expressed as, Equation 6 is shown as:

$$e_{ij}^{\epsilon} = \,\frac{1}{2}\left(\frac{\partial u_i^{\epsilon}}{\partial x_j} + \frac{\partial u_j^{\epsilon}}{\partial x_i}\right) \tag{6}$$

Leading to the stress tensor being expressed in Equation 7 as:

$$\begin{aligned} \sigma_{ij}^{\epsilon} = \quad & C_{ijkl}\,\frac{\partial u_k^{\epsilon}}{\partial x_l} \\ & = C_{ijkl}\left(\frac{1}{\epsilon}\frac{\partial u_k^{(0)}}{\partial y_l} + \left(\frac{\partial u_k^{(0)}}{\partial x_l} + \frac{\partial u_k^{(1)}}{\partial y_l}\right) + \epsilon\,\frac{\partial u_k^{(1)}}{\partial x_l}\right) \\ & = \frac{1}{\epsilon}\,\sigma_{ij}^{(-1)} + \,\sigma_{ij}^{(0)} + \epsilon\sigma_{ij}^{(1)} \end{aligned} \tag{7}$$

And finally using Equation 7 to create the equilibrium equation shown in Equation 8:

$$\frac{1}{\epsilon^2}\frac{\partial \sigma_{ij}^{(-1)}}{\partial y_j} + \frac{1}{\epsilon}\left(\frac{\partial \sigma_{ij}^{(-1)}}{\partial x_j} + \frac{\partial \sigma_{ij}}{\partial y_j}\right) + \left(\frac{\partial \sigma_{ij}}{\partial x_j} + \frac{\partial \sigma_{ij}^{(1)}}{\partial y_j}\right) = b_i \tag{8}$$

This creates three separate equations for use with Finite Element Method (FEM), with the boundary conditions of the unit cell shown in Equation 9 (with u_i^1 being equal at the interface of the two length scales), resulting in Equations 10, 11, and 12:

$$\frac{\partial \sigma_{ij}}{\partial y_j} = 0 \tag{9}$$

$$\frac{\partial \sigma_{ij}^{(-1)}}{\partial y_j} = 0 \tag{10}$$

$$\frac{\partial \sigma_{ij}^{(-1)}}{\partial x_j} + \frac{\partial \sigma_{ij}}{\partial y_j} = 0 \tag{11}$$

$$\frac{\partial \sigma_{ij}}{\partial x_j} + \frac{\partial \sigma_{ij}^{(1)}}{\partial y_j} = b_i \tag{12}$$

FEM was used, with a 3 dimensional, 8 node hexahedral element (Li & Wang, 2008), (Fish & Belsky, 1995), where N_A is the tri-linear shape function:

$$u^* = \sum_{A=1}^{n_{eq}} N_A \boldsymbol{d}_A \tag{13}$$

$$\delta u^* = \sum_{A=1}^{n_{eq}} N_A \delta \boldsymbol{d}_A \tag{14}$$

Leading to the discretized unit cell problem being expressed in Equation 15 as:

$$Kd = f \tag{15}$$

With Equation 16, 17, and 18:

$$K_{AB}^e = \int_{\Omega_e} B_A^T D B_B \, d\Omega \tag{16}$$

$$f_A^e = -\int_{\Omega_e} B_A^T D \bar{\epsilon} \, d\Omega \tag{17}$$

$$B_A = \mathcal{L}(N_A) = \begin{bmatrix} \frac{\partial(N_A)}{\partial y_1} & 0 & 0 & \frac{\partial(N_A)}{\partial y_2} & \frac{\partial(N_A)}{\partial y_3} & 0 \\ 0 & \frac{\partial(N_A)}{\partial y_2} & 0 & \frac{\partial(N_A)}{\partial y_1} & 0 & \frac{\partial(N_A)}{\partial y_3} \\ 0 & 0 & \frac{\partial(N_A)}{\partial y_3} & 0 & \frac{\partial(N_A)}{\partial y_1} & \frac{\partial(N_A)}{\partial y_2} \end{bmatrix} \tag{18}$$

Thus, the macro-scale material properties matrix will be shown in Equation 19:

$$\boldsymbol{D} = \frac{E}{(1+\nu)(1-2\nu)} \begin{bmatrix} 1-\nu & \nu & \nu & 0 & 0 & 0 \\ \nu & 1-\nu & \nu & 0 & 0 & 0 \\ \nu & \nu & 1-\nu & 0 & 0 & 0 \\ 0 & 0 & 0 & \frac{1-2\nu}{2} & 0 & 0 \\ 0 & 0 & 0 & 0 & \frac{1-2\nu}{2} & 0 \\ 0 & 0 & 0 & 0 & 0 & \frac{1-2\nu}{2} \end{bmatrix} \tag{19}$$

To determine the D matrix for the mycelium materials, the mechanical properties must be defined for both the matrix (the mycelium cellular foam), and the heterogeneous fibrous reinforcement (the undigested wood fibers). Using the theoretical Young's Modulus of mycelium determined by (Van Suijdam et al., 1982) as 20.0 MPa, and solving using an inverse problem method results in determining the following results. With Poisson's ratio of wood 0.38 (at 12% humidity), and Poisson's ratio of mycelium foam, 0.25 and Young's Modulus of wood 1200 MPa (Green at al., 1995), and the homogenized modulus of mycelium noted above as 20.0 MPa - the method predicted the Young's Modulus of mycelium materials to be calculated as 0.96 MPa. Comparing the predicted mycelium foam's elastic modulus and the experimentally determined modulus during this research to be 1.30 MPa, with a difference in values around 35%, the asymptotic homogenization method can be considered suitable. As both the predicted and experimentally determined modulus were in the order of 1 MPa, the mycelium materials are suited to lower-load applications that do not require high material stiffness. To improve the mycelium materials properties, they can be combined with higher-strength materials as a composite, if stiffness is a required property. Mycelium materials can be considered as similar to synthetic foams containing wood fibers, lending insight into how the cellular properties could change the mycelium materials properties. Khazabi et al. (2011) found that wood fibers in a polyurethane spray foam increased cell stiffness. Cells also became larger and more irregular with increasing fiber concentration, and the foam became less homogeneous. Shorter fibers led to a higher number of cells with decreased cell wall thickness. Increasing the proportion of wood fiber in the polyurethane foam increased the compressive strength and decreased the tensile strength. They attribute these results to both the stiffening of the cell walls, as fibers are incorporated, as well as the mutilation of cell shape due to fiber interference. These studies considered fibers up to 0.04 inches (1.0 mm) - slightly larger than the foam cell size (Khazabi et al., 2011). If considering mycelium materials are similar, being a cellular matrix with fibers, smaller feedstock fibers should result in a material with more regular cells and a higher strength material. In Holts' (Holt et al., 2012a) study of mycelium grown on cotton plant material, no clear relationship was established between feedstock particle size and resulting mechanical properties. Similar studies have found wood fiber reinforcements have an impact on elastic and mechanical properties of the resulting composite. Elastic properties typically increase with hardwoods, such as Red Oak feedstock used here, and also with defibration (the separation of fibers from one another), when macerated into wood chips (Marklund et al., 2008). Of the varied types of mycelium materials that have been seen during this research, despite lacking enough materials to empirically test, anecdotal evidence has been that a finer feedstock (and thus finer reinforcing fibers) result in a finer material consistency and harder, denser, and stronger material. Should more materials be available, this would be a key area to continue research in.

Chapter Two Conclusion: Mycelium Composites, a Cellular Material

To satisfy the need for empirically determined material properties, and to understand the structure and resulting physical properties of the mycelium-based materials, the density was experimentally determined. Optical & scanning electron microscopy identified mycelium growth as a white colored mass of hyphae, concentrated around the feedstock chips, and gave some insight into the frangible nature of the material, which occurs at the interface of the mycelium and feedstock chips. To address frangibility in future research, the ingress via digestion and adhesion of hyphae to the feedstock could be investigated. Scanning electron microscopy identified the random, tangled nature of the hyphae growth, showing a structure closer to a synthetic fiber random-mat structure, than the theorized cubic open-walled cell structure. This opens further avenues of research to determine if the random structure results in isotropic material properties, and also lends insight into the insulation properties of mycelium materials due to the air spaces in-between the hyphae that prevent effective heat transfer. The scanning electron microscopy images were also used to identify the diameter of an individual hypha, which was found to be in the range of 0.5-1.0 µm, similar to previously reported diameters in the range of 2.0-3.0 µm (Bhosle et al., 2010), (Li et al., 2015). Scanning electron microscopy also noted the formation of a continuous layer of material at the interface of the living material with physical boundaries, including the air space above the growing material. This suggests the morphology and structure of the material could be controlled by controlling the growing conditions and the growing space. For example, higher density materials could be formed by inducing multiple internal stacks of the continuous layer, or conversely by artificially removing the boundary conditions to encourage material growth without continuous layer boundaries. This would be most applicable to the possibility of joining smaller growth blocks into a larger structure, and allowing the fusing of the blocks through natural growth, as an alternative to requiring adhesives.

Experimental measurement identified the material density as 318 kg/m^3, with the difference between previously published and experimental values being due to the natural variation of the material formed resulting from varied growth periods, as longer growth periods result in more complete digestion of the feedstock and thus higher density. This suggests the final density can be controlled both through controlling the growth time and the rate of digestion. Based on an asymptotic homogenization method and empirical testing, the Youngs Modulus was found to be 1.30 MPa. This suggests the material is suited to applications that don't require high stiffness, or combined with other, stiffer materials as a sandwich composite. Overall, mycelium materials were shown to be a light-weight, low-density, naturally grown material, able to take a variety of forms through the controlled growth. The structure is formed of millions of individual hyphae, which can be modeled with an asymptotic homogenization designed for the resulting repeating cellular materials, modeled an open cell cube but found to shows a random fiber mat-like structure. This favors applications where low thermal transmission is a useful property, such as insulation, and light-weight, low strength applications such as packaging. The range of properties of the mycelium materials suggests good potential for further investigation through empirical testing, to determine the mechanical properties of various types and densities of mycelium material such as explored by Allwood et al. (2012), which would help inform researchers, designers, and provide a reference point when finding suitable applications.

Chapter Three – Determining Strength, Stiffness & Other Material Properties

Development of novel materials, in particular natural ones, requires a high degree of innovation. Multiple aspects of a material have to be explored, to ensure the properties fit the required specification, which is further complicated by the biological growth processes and resulting variation that are intrinsic to natural materials. Testing standards and procedures specifically for biomaterials are also still being fully established alongside bodies of technical work. Testing novel natural materials also experiences a high degree of variability in results in comparison to synthetic materials (Ashby & Johnson, 2013), as the very nature of growing materials – which digest their feedstock and convert it into their structure – means there is a high degree of internal variation within the material itself. This can be due to variation in the feedstock, itself being a natural product, and variation in the response to growing conditions. The following mechanical characterization of the mycelium materials was carried out based on ASTM testing standards. While no specific mycelium-based material standards exist, polymer foam and composite standards were selected as the most appropriate, due to having similar characteristics. Determining the tensile and compressive yield strength, and tensile and compressive ultimate strength, and the Young's Modulus of the material will help inform researchers and commercial developers about the material performance. This will help inform the comparative mechanical properties of mycelium compares to similar materials, and help inform developers of suitable applications.

Tensile & Compressive Testing Methods and Results

Tensile and compressive tests were conducted to determine the mechanical behavior of the mycelium material under static tensile and compressive loads. Tensile and compressive tests were carried out according to ASTM D3574 – 11 Standard Test Methods for Flexible Cellular Materials—Slab, Bonded, and Molded Urethane Foams (since superseded by ASTM D3574-17 in 2017) (ASTM D3574, 2017), chosen as the most appropriate standard for testing mycelium composites. ASTM D3574 is normally used for polyurethane foam, however this standard provides the most applicable testing methodologies for a natural material with a foam-like structure, as no testing standards currently exist specifically for natural composite foams. Any adaptations to the methods or test fixtures are noted to help inform future research efforts.

Samples of *Ganoderma lucidum*-based mycelium materials were selected, with further details on the fabrication of the material given in Chapter 9. The samples were cut into test specimens using a General Electric vertical band saw at 1725 rpm. This proved difficult due to the frangible structure of the material, and considerable debris was produced during cutting. During test specimen fabrication, it was noted the material was brittle and frangible, with fracture occurring at the boundaries between areas of mycelium growth and undigested wood chips, implying a weak bond. Compressive and tensile specimens were fabricated according to ASTM D3574, although some adaptations had to be made due to the frangibility of the material. The tensile samples 'dogbone' shoulder radii were replaced with a chamfer to aid in fabrication, and the resin-glued metal tabs with an 8mm diameter centered blind hole were extended beyond the sample to enable fitment into a pin-clamp, as standard crush-jaw clamps would damage the specimens. The tensile samples had an average mass of 40.9 g, of dimensions 25.2 x 25.7 x 141.1 mm, and a moisture content of 5.7%. The compression samples had an average mass of 40.7 g, of dimensions 55.1 x 55.4 x 41.9 mm, and a moisture content of 5.8%. Mass, dimensions and moisture content were recorded for each specimen, and the average determined for subsequent use in calculations. Due to the mycelium material being desiccated, the moisture content of the material is ~6%, which is less than 10% of the surrounding environmental humidity. Therefore it was noted that the material is mildly affected by the surrounding environments moisture content, including air humidity, so the moisture content was recorded at the time of testing using a digital resistivity moisture meter with an accuracy of 1%. Due to the lack of available materials, the number of tests were limited and thus the following method was selected to result in the highest possible accuracy. Of the three tensile and three compressive tests planned, all six tests were completed successfully. Load deflection plots of the materials response to tensile and compressive loads were recorded during each test, averaged, and the resulting stress-strain responses plotted.

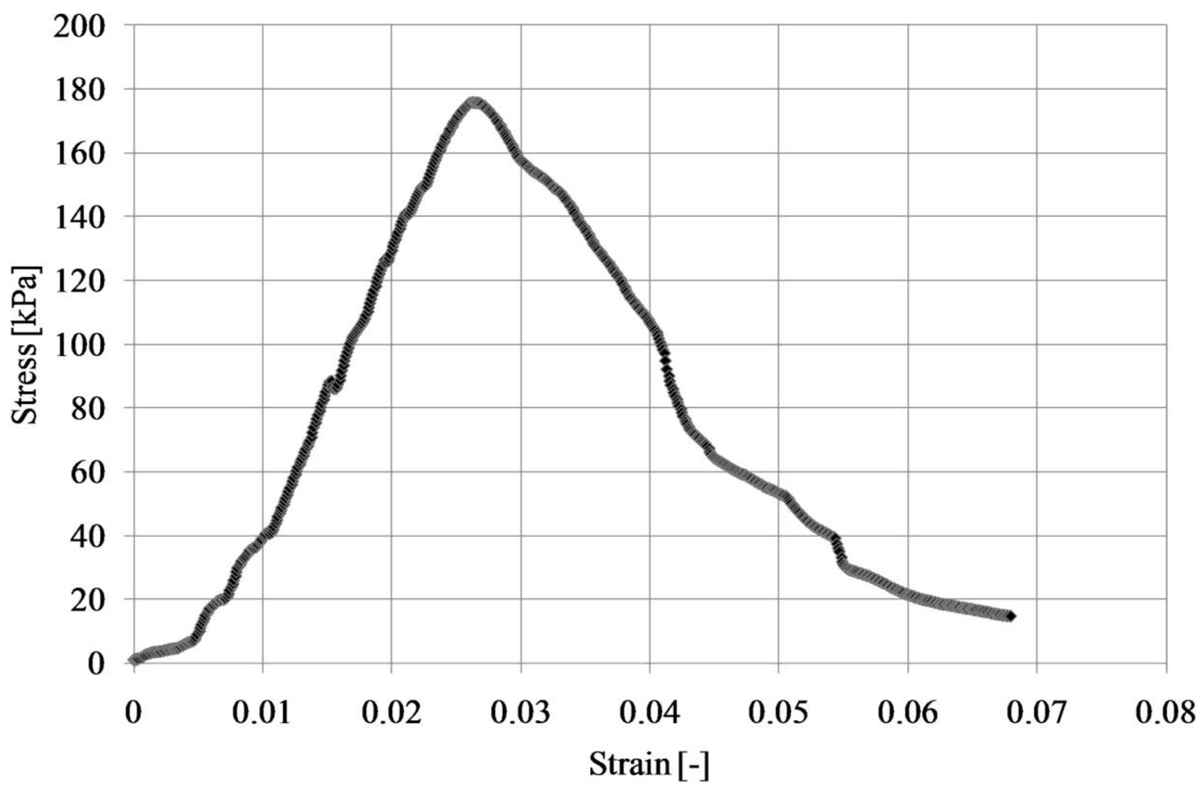

Figure 12. Tensile test stress-strain response

The stress-strain response in Figure 12 of the material under a tensile load was seen to follow the expected pattern for an elastic-plastic material: a linear-elastic region up to a 0.2% offset yield point of 15 kPa, a negligible plateau region, and finally plastic deformation until brittle fracture at 176 kPa. The stiffness (Young's Modulus) of the material in tension, calculated from the elastic extension portion of the graph was found to be 1.3 MPa. The relatively low Ultimate Tensile Strength (UTS) of 176 kPa reflects the brittle and frangible nature of the material. While a relatively low UTS compared to synthetic materials, however for current applications such as packaging, the material is typically not be subject to tensile loading. However, additional reinforcements, and consolidation to reduce frangibility, may improve the tensile properties if this is necessary.

The compressive test curve Figure 13 includes the three regions of linear elasticity, plateau and densification. At very low values (~≤5%) the materials response is linear elastic, where the gradient equates to the compressive stiffness of the mycelium material. As expected for a foam matrix, deformation was followed by crushing of the foam cells, in this case the struts of the mycelium hyphae architecture, with densification seen as a sharp increase of stress to strain. As load increases, the struts of the cell walls begin to collapse. The collapse of the cells and thus material, continues approximately constantly until the opposing cell walls meet, causing the stress response to increase sharply, identified as densification (Han et al., 1998), (Avalle et al., 2001). The material stiffness in compression was calculated as 1.0 MPa, using the elastic, proportional region of the graph in Figure 13 prior to the compressive yield at 47.5 kPa. The ultimate compressive strength of 490 kPa proved considerably higher than the UTS.

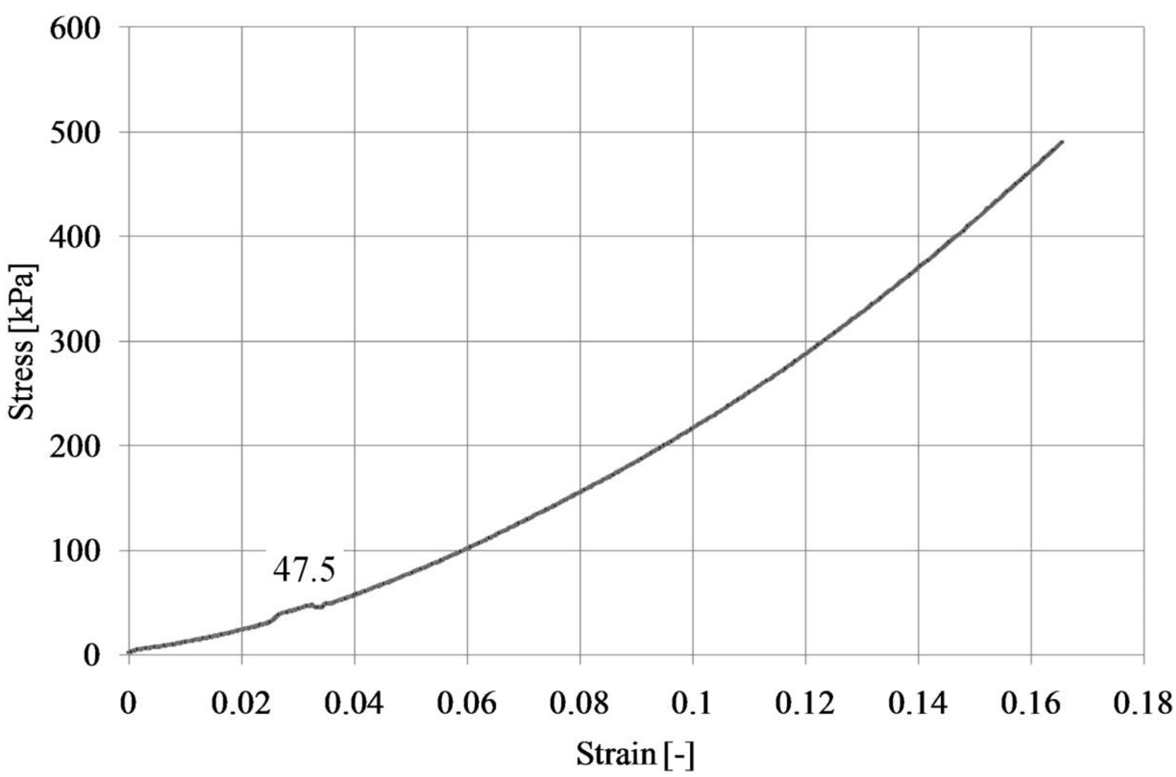

Figure 13. Compressive test stress-strain response

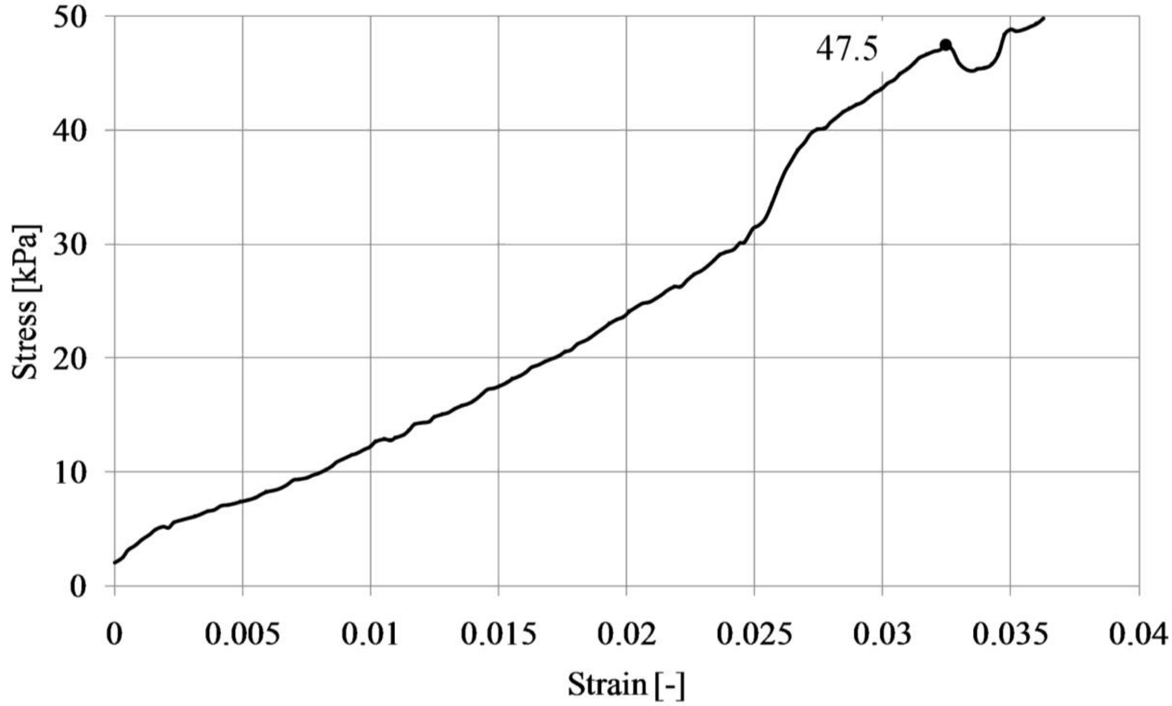

Figure 14. Yield point of compressive stress-strain response

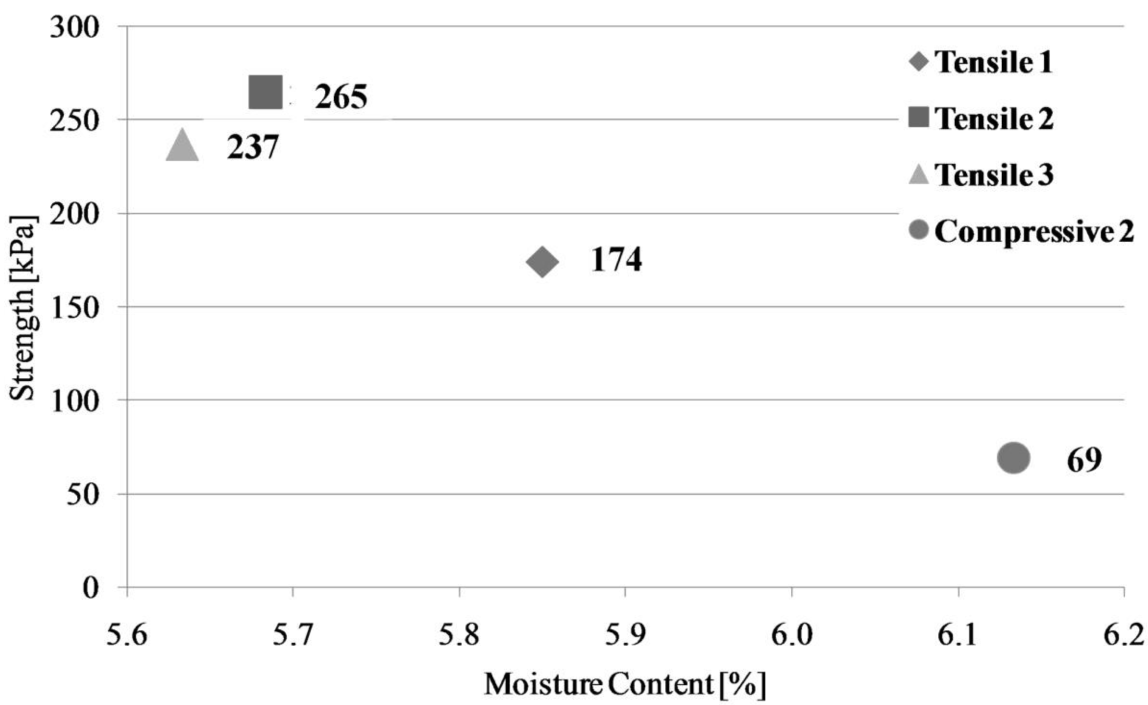

Figure 15. Mycelium material strength in relation to moisture content

The material strength was found to decrease with increasing moisture content of the material, as seen in Figure 15. This may be due to moisture affecting the material bonds, although this require further investigation to confirm. The conclusion is that the moisture content reduction process during material fabrication is vital to ensure consistency and performance, and that coatings to inhibit moisture diffusion could improve performance, especially during the material use phase.

The predicted modulus of the mycelium foam material was calculated, by dividing the mass by volume, with the density of dried mycelium as 1400 kg/m^3 and modulus of the mycelium theorized as 20.0 MPa (Van Suijdam et al., 1982).The experimental measured tensile modulus was 1.30 MPa. The difference between the literature and model predicted modulus, and the experimentally measured modulus, implies the importance of accounting for the density and modulus of the undigested wood chips present in the mycelium. If considered as a composite material of remaining wood chip clusters as fiber reinforcements and the grown mycelium as matrix, an estimation of the composite density could be made using the rule of mixtures. Since the mass fractions of fiber and matrix cannot be found from the raw materials due to the conversion of the carbon-containing cellulose into mycelium cellular structures, the composite density could only be calculated by estimating the wood fiber and mycelium matrix volume fraction ratio. Currently there is no established method to determine the ratio of mycelium matter to wood chip feedstock matter in natural composites. Several methods to estimate the relative composition fraction of wood and mycelium were considered, including estimation by analysis of SEM image or visual inspection, but neither method was developed further or tested for accuracy.

Mycelium Materials Compared to Natural & Synthetic Contemporaries: A Natural Foam

In summary, the mycelium material was seen to be a composite material of mycelium foam matrix and wood reinforcement. No growth orientation was found, and SEM images confirmed that the mycelium matrix formed around the wood chip regions, forming a network of hyphae that shows increasing density towards the self-skinning exterior of the material. Static mechanical tests showed the frangible nature of the material resulted in a low material ultimate tensile strength of 176 kPa, with a tensile modulus of 1.30 MPa. In compression the material demonstrated a yield point of 47.5 kPa, ultimate compressive strength of 490 kPa, and compressive stiffness of 1.0 MPa.

This shows the material is better suited to applications that require compressive strength, as opposed to tensile strength, as the mycelium material exhibited a compressive strength almost three times the tensile strength. The mechanical properties of the mycelium materials are summarized in Table 2. When compared to commonly used polymer foam materials, the mycelium material had an average density and strength comparable to polymer foams, indicating properties closest to expanded polystyrene foam. While the mycelium material is not comparable to the mechanical performance of metal foams, in relation to polymer materials, mycelium material have the advantage of being naturally occurring, sustainably produced, and free from toxic manufacturing process including toxic chemicals present in polymer foams.

Material	**Modulus, E (MPa)**	**Density, ρ (kg/m^3)**	**E / ρ**	**Yield Strength, S_{yc}**	**Ultimate Strength, S_{utc}**	**S_{yc} / ρ**	**S_{utc} / ρ**

				(MPa)	(MPa)		
Ganoderma lucidum (Present investigation)	1.30	318	0.004	0.0475	0.176	0.0001	0.002
Starch-based foam (w/ fiber)	183	260	0.70	1.18	1.09	0.005	0.004
Polystyrene foam	5.70	41.2	0.14	-	0.18	-	0.004
Polyvinyl-chloride foam	3.00	500	0.06	-	45.0	-	0.900
Aluminum foam	347	255	1.40	1.69	-	0.007	-

Table 2. Comparative Ganoderma lucidum material properties (Polat et al., 2012), (Sadak & Fouad, 2013), (Wilkes er al., 2005), (Ramamurty & Paul, 2004), (Ashby & Medalist, 1983), (Stevens et al., 2010).

Natural materials are typically lower strength than costly synthetic materials. With lower strength natural materials, cellular structures are also common, with parenchyma tissues commonly found in nature as sugar storage within plants such as potatoes, which also form a polyhedral cellular structure explained by Gibson as being formed of a pressurized, liquid-filled closed-cell foam (Gibson, 2012). Higher modulus and stiffness natural materials also exist which result in greater strength, typically also in cellular formats such as soft woods, as seen in Figure 18, with cellulose being able to achieve a strength of around 1000 MPa, which is a similar to synthetic composites.

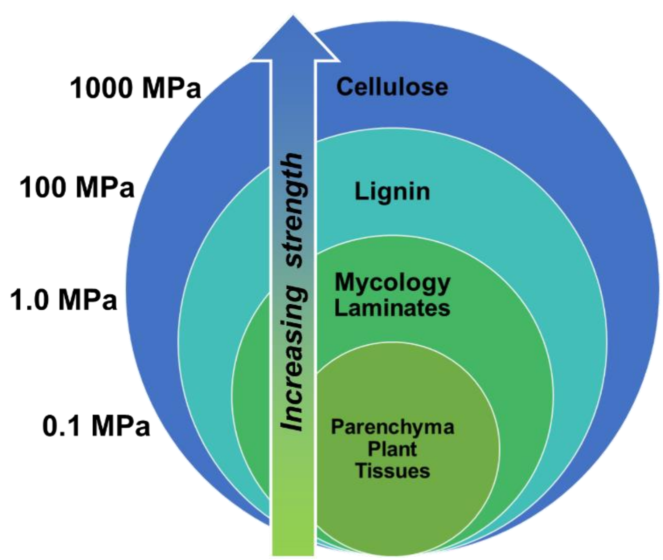

Figure 16. Natural materials relative order of magnitude of tensile strengths. Adapted from (Gibson, 2012)

Holt et al. (2012a) further described material processing and properties, using a feedstock composed primarily of cotton carpel (a waste part of cotton plants), along with smaller proportions of cotton seed hulls, starch, and gypsum. Their study compares twelve different growth methods, using six different ranges of particle size for the carpel, from 0.1-51.0 mm, and two methods for inoculation via a liquid or a solid substrate. They found the liquid substrate was easier to implement and also resulted in more uniform delivery (Holt et al., 2012a). The feedstock blend was heated for sterilization, inoculated with a *Ganoderma lucidum* fungus, packed into a mold, and sealed to create a uniform environment. Mycelium growth occurred over five days at room temperature, after which the material was held at a higher temperature to stop growth (Holt et al., 2012a). The study measured a range of properties, using primarily ASTM published testing standards. Their results for mechanical properties are summarized in Table 3. Despite the mechanical properties showing a very wide range, Holt et al. concluded that mycelium presents a practicable alternative to polystyrene packaging foams (Holt et al., 2012a), which the mechanical characterization during this research supports as an application for lower strength, environmentally lower-impact materials.

Density (kg/m^3)	**Flexural Strength* (kPa)**	**Elastic Modulus* (kPa)**	**Compressive Strength* (kPa)**
66.5-224	7-26	123-675	1-72

*normalized to polystyrene density $32 kg/m^3$

Table 3. Properties of mycelium materials grown from cotton carpel feedstock (Holt et al., 2012a)

While the properties of mycelium materials make them comparable to polymer foams and lower-strength natural materials such as cork, their manufacturing, environmental, and other abilities often provide advantages over such materials. For example, while mycelium materials are similar in mechanical performance to expanded polystyrene, they provide much less environmental impact due to being recyclable through composting. Defining and comparing these qualities quantitatively can be problematic, due to the lack of available information, and also the lack of context and intuitive meaning when comparing data points. To solve this, the following Table 4 aims to provide a comparative guide to the performance of mycelium materials, in relation to similar synthetic and natural materials, using a relative low-to-high rating scale. Different rating may or may not be advantageous depending on the material and property, for example a 'low' density is advantageous for mycelium and polystyrene foam (such as for packaging applications), but a 'low' composability is not advantageous for the same materials. To address this, a color code has also been applied, to indicate the rating of being advantageous (green), medium (yellow), or not advantageous (pink).

	Mycelium	**Mycelium sandwich composite**	**Balsa wood**	**Natural cork**	**Polystyrene foam**
Density	Low	Medium	Low	Low	Low
Tensile strength	Low	Medium	Medium	Low	Low

Compressive strength	Low	Medium	Medium	Low	Low
Shear strength	Low	Medium	Medium	Low	Low
Tensile stiffness	Low	Medium	Low	Low	Low
Frangibility	High	Low	Low	Medium	Medium
Degradation due to H_2O	High	High	Medium	Medium	Low
Mechanical cutting performance	Low	High	High	High	High
Thermal laser cutting performance	High	Low	High	High	High
Ability to form small features (~5.0 mm)	Low	Medium	High	Low	Medium
Maximum temperature	Medium	High	Medium	Medium	Low
Ability to form small features (~5.0 mm)	Medium	Medium	High	Medium	Medium
Composability	High	High	Medium	Medium	Low
Manufacturing complexity	Low	Low	Low	Low	High
Energy content	Low	Low	Low	Low	High

Table 4. Mycelium materials comparative performance guide

Considering the entire range of properties, mycelium materials cannot achieve the mechanical and manufacturing performance of a synthetic polymer, but can offer better environmental properties. Communicating these advantages to designers will help increase adoption of mycelium material for applications requiring less impactful environmental choices, but not requiring high strength.

Mechanical Properties Conclusions

Mechanical testing identified the mycelium materials as having an ultimate tensile strength of 176 kPa, with a tensile modulus of 1.3 MPa, suggesting the frangible nature of the material resulting in comparatively low strength. The yield point was identified as 47.5 kPa, and the ultimate compressive strength of 490 kPa. The compressive strength of the material was found to decrease with increasing moisture content of the material, thus the moisture content reduction process is vital to ensure consistency and performance. Due to achieving an ultimate compressive strength around three times the ultimate tensile strength, mycelium materials are determined to be suited to compressive applications, such as packaging, where the low strength of the material under tensile loading does not impact performance. This also suggests mechanical performance could be improved by further investigation to various composite formations, such as sandwich composites which could provide higher mechanical performance via various material combinations.

Chapter Four – Thermal Characterization

Due to their cellular structure and natural composition, mycelium materials make an attractive option for insulation applications. As they can be faced with either natural or synthetic laminate materials, they also have good potential as a natural composite insulation panel for use in construction applications. The thermal insulation properties of mycelium materials were characterized to provide information for their development into such applications. To this end, the properties useful in determining the suitability of a mycelium material being used for a thermal insulating application were investigated using ASTM testing methodologies.

Heat Resistance: Determining the Maximum Use Temperature

Determining the maximum temperature that mycelium materials can be used within is a key property designers and developers need to know, when considering these materials for various applications. The maximum use temperature of the material is determined using ASTM C447 − 03 (Reapproved 2010) Standard Practice for Estimating the Maximum Use Temperature of Thermal Insulations (ASTM C447, 2013). Determining this property is important to understand the applications it can be used for, such as performance when next to heat sources, such as when in close proximity to heat sources in kitchen environments, or next to heating and ventilation equipment. Maximum use temperature is a key property designers and engineers need to know when determining if a material can meet the needs and specification of the application they are considering using the material for.

The test evaluates the dimensions, weight and appearance of the insulation material measured before, during, and after exposure to a hot surface, as according to ASTM C447. The dimensions, weight and surface conditions were described and evaluated pre- and post- application to a hot platen face at temperature intervals of +122ºF (+50ºC) beginning from 68ºF (20ºC). The assessment of the materials performance was carried out according to ASTM C411-17 Standard Test Method for Hot-Surface Performance of High-Temperature Thermal Insulation (ASTM 411, 2017). Any notable sag and warpage will be noted prior to the tests. Post-tests any sag, warpage, cracking, delamination, flaming, glowing, smoldering, or smoking will be evaluated and reported, and the temperature of the occurrence of these found to be the maximum use temperature of the mycelium materials.

The dimensions of the test specimens did not appreciably change during the test. The weight of the test specimens was measured before and after the test, with all specimens reducing in weight by an average of 2.5%. This matches with the reduction of moisture content measured before and after the test. Prior to testing the average moisture content was 10.7% and after the test the average moisture content was reduced to 10.3%, and both are attributed to the drying effect of exposure to the hot surface reducing the amount of water moisture in the materials. Before the test, no test specimens had notable sag, warpage, cracking, or delamination. A full list of materials types tested is given in Table 5, and summarized as materials types A – M (except D & J) being various mycelium materials with no laminate skins and generally decreasing density, and materials types D & J being mycelium materials with bamboo and plywood laminate skins.

Each test specimen was applied to the hot surface at increments of +50ºC (122ºF). After exposure to the hot surface, the materials showed a range of evidence of smoldering, the beginning stage of combustion of the material. Material types M and E showed evidence of smoldering at 450ºC (842ºF), close to the maximum use temperature of cellular glass. Material types B1, B2, D, G, I, L, showed evidence of smoldering at 400ºC (752ºF), and types A, C and J and K, showed evidence of smoldering at 350ºC (662ºF). Material H showed evidence of smoldering at 300ºC (572ºF), as visualized in Figure 17 below.

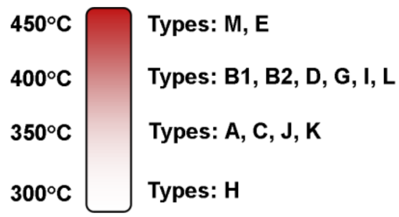

Figure 17. Mycelium materials types and maximum use temperature

No clear correlation was found between maximum use temperature and density, suggesting resistance to inflammability is not related to internal void content but rather the material composition. All material types thermally degraded by smoldering, a low temperature combustion of the material as fuel by oxygen. This is the same process that occurs to cellulose, wood and polymeric polyurethane foams (Ohlemiller, 1986). Examples of the materials pre-and post- test are shown in Figure 18 below, with mycelium material type L showing discoloration and charring, unevenly distributed due to the uneven surface of the material. Material type J, faced with bamboo laminates, showed more charring, likely more evenly distributed due to being a flatter surface. Both represent common options for commercial applications of mycelium materials, and thus were chosen to test determine the maximum use temperature.

Figure 18. Material types L and J before exposure to hot surface in 1 and 2 respectively, and after in 3 and 4.

The safe material temperature range, defined as use temperatures which result in no evidence of heat-based degradation, except slight surface discoloration, was identified as between 68ºF (20ºC) to 662ºF (350ºC), with the average temperature of maximum use calculated as being 734ºF (390ºC). As above 572ºF (300ºC) one type of mycelium material was found to suffer discoloration and eventual smoldering, thus the maximum use temperature was found to be under 572ºF (300ºC). The maximum use temperatures of mycelium materials were found to be higher than polyurethane and phenolic foams, and more than double the maximum use temperature of polystyrene foam. This shows an opportunity for mycelium materials to be developed as replacements for synthetic foams, many of which are thermoplastic polymers which melt at relatively low temperatures. Further mechanical testing of the mechanical properties, such as the compression set, would be required to evaluate their mechanical suitability at elevated temperatures. Despite this, mycelium materials thermal properties show promise for suitability for a range of thermal insulation applications. With the benefits of being a non-toxic and compostable, reducing environmental impact, mycelium materials are a good option for a natural insulating material such as for use in structural insulating panels, and merits further development and investigation.

Mycelium Materials as a Natural Insulator

The lightweight properties of mycelium materials led commercial developers to explore mycelium materials as insulation. Synthetic insulations such as polymer foams and glass fiber, while being lightweight, pose dangers to environmental and human health. For example, urethane foam insulation emits formaldehyde vapors, which are distasteful but not dangerous if properly handled (Thomas, 1990). Glass fibers are also hazardous to health in various formats (Steenland & Stayner, 1997), leading to additional cost and time needed to safely handle such materials including personal protective equipment. Due to this, finding natural alternatives for insulation materials, particularly lightweight and environmentally sustainable ones, has been a priority of commercial developers of mycelium materials. The cellular and air pocketed structure of the mycelium materials lend themselves to insulating applications, due to the high ratio of air space to solid material and cells being in the micro-range of size, minimizing heat transfer through air convection and conduction. Due to these insulating properties, there is potential for using mycelium materials for thermal insulation applications, such as Structural Insulated Panels (SIPs), a composite building material with an insulating core layer (often of polystyrene) sandwiched between two layers of structural wood board. Since the mycelium materials can be grown to produce lightweight, low density, and flame resistance properties, and can be considered as an alternative to polystyrene, mycelium materials present themselves as an attractive choice for evaluation and characterization of insulation properties and suitability for such applications. Consumers may also find a 100% recyclable material, made from sustainable feedstocks of waste agricultural materials, an attractive alternative to toxic and crude oil-based insulating materials used in their living spaces.

Determining the effectiveness of the mycelium materials as a thermal insulation material is a key part of supporting their development for commercial applications. One of the most important properties of insulation materials is their ability to reduce transmission of heat flux from a hot surface, for example the warm indoor environment, to a cold surface such as a building's exterior wall. Performance as an insulation material is measured in a materials ability to reduce the transmission of heat flux, Q, from a hot surface to a cold surface. The efficiency of insulating materials is described in industry as the R value, a measure of the thermal resistance of specimen in units of m^2 Kelvin per Watt. This value will be experimentally determined for each type of mycelium material. To determine the transmission of heat flux, a hot surface and cold surface were used to measure the heat flux across a materials test specimen. Testing was informed by ASTM C1044–12 Standard Practice for Using a Guarded-Hot-Plate Apparatus (ASTM 1044, 2012). The testing methodology used a single sided thin heater hot plate, insulated to prevent lateral and edge heat losses. The temperatures of the hot and cold surfaces were measured with thermocouples, as according to ASTM C177 – 13 Standard Test Method for Steady-State Heat Flux Measurements and Thermal Transmission Properties by Means of the Guarded-Hot-Plate Apparatus (ASTM C177, 2013), as specified by the ASTM C1044 methodology. Measurements were made once the set test temperature was achieved, at the point of steady state conditions. The hot plate contact area, A in m^2, heat flow, Q in Watts, surface temperature of hot surface Th in ºK, surface temperature of cold surface Tc in ºK, will be recorded, along with the specimen thickness, L, in m. Following the test the R value in m^2 K/W, the thermal resistance of specimen will be calculated according to C1044–12, using Equation (20), and the thermal conductivity, C, in W/m K will be calculated using Equation (21).

$$R = (A\,(T_h - T_c))\,/\,Q \tag{20}$$

$$C = (\mathrm{L}Q)\,/\,(A\,(T_h - T_c)) \tag{21}$$

The effectiveness of the mycelium materials as an insulation material, the R value, the thermal resistance of the mycelium materials in units of m^2 Kelvin per watt were found using ASTM C1044–12 Standard Practice for Using a Guarded-Hot-Plate Apparatus at 396 ºK. Once the point of steady state temperatures was achieved, the hot surface temperature Th in ºK, and cold surface temperature of Tc in ºK, were measured. Using equation (20), the R value, the thermal resistance of specimen in units of m^2 K/W. Using equation (21), thermal conductivity, C, in W/m K were calculated and reported in Table 5 for each material type, along with each materials' density. Fourteen different types of mycelium materials were tested, specified in Table 5. Specimens were uncoated and left in their natural finish.

Testing established the following values; the thermal resistance, R, ranged from 0.0014 to 0.0019 m^2 K/W, and values of thermal conductivity, C, ranged from 0.053 to 0.077 W/m K, as seen in Table 5 below. This is comparable to balsa wood (0.048 W/m K) and natural cork (0.070 W/m K) (EngineeringToolbox, 2018). For comparison to synthetic materials, fiber glass has a thermal conductance of 0.040 W/m K (EngineeringToolbox, 2018). This means that mycelium materials would be a suitable replacement for natural cork, which is a well-established natural material in the architectural industry, but has limited natural supplies and causes environmental impact due to being transported long distances. Mycelium materials would also be a suitable replacement for glass fiber insulation, and since they can be produced in large volumes from sustainable sources, also indicating good potential for further development.

Material Type	Density (kg/m^3)	C (W/m K)	R (m^2 K/W)
Type A	410	0.057	0.0015
Type B1	364	0.059	0.0015
Type B2	344	0.066	0.0017
Type C	315	0.062	0.0018
Type D	432	0.057	0.0016
Type E	715	0.072	0.0015
Type F	773	0.061	0.0019
Type G	347	0.077	0.0015
Type H	564	0.056	0.0014
Type I	321	0.065	0.0017
Type J	428	0.063	0.0023
Type K	407	0.069	0.0014
Type L	390	0.057	0.0018
Type M	359	0.053	0.0016

Table 5. Mycelium materials type, density, thermal resistance (R) and thermal conductivity (C).

The favorably low thermal conductivity values of mycelium materials may be due to the high proportion of air in the material, which has a cellular structure and is modeled as an open-walled cellular foam. Similarly, cellular wood materials, which often are the feedstock for mycelium materials have in-built voids of air that insulate heat transfer. As the density of air decreases as temperature increases, as the materials temperature increases it reduces heat conduction occurring through the air voids (Suleiman et al., 1999), aiding the insulation abilities of the material.

The mycelium materials of *Ganoderma lucidum* with more complex feedstocks and higher density resulted in higher values for thermal conductivity, and *Ganoderma lucidum* materials with lower density resulted in lower values for thermal conductivity. The lamination of the *Ganoderma lucidum* core with bamboo and ply laminates, in types J and D respectively, did not seem to significantly affect the thermal conductance either positively or negatively. Three results did not correlate as closely, for material type material E, F and J. This was possibly due to materials type E and F have a comparatively high density of over 700 kg/m^3, in comparison to the other mycelium materials. Material type J, being a bamboo laminated sample, achieved a thermal resistance of 0.0023 m^2 K/W, which follows sandwich composites having insulation properties due to multiple material layers. However, the balsa wood laminated sample, material type D, achieved 0.016 m^2 K/W, potentially attributing type J's insulation to the bamboo material, rather than the sandwich composition. This may be due to bamboo's naturally high resistance to heat flux, with C values of up to 0.35 W/m K, again due to the highly cellular structure of the material (Shah et al., 2016). Had there been more material available, further testing to determine the thermal properties of the bamboo and balsa wood individually would provide more information & clarity on mycelium sandwich composites thermal behavior, however time and resources did not allow for this.

To investigate the relationship between the mycelium material density and the thermal resistance (the commonly used value to describe a materials ability as a thermal insulator), a scatter graph was plotted of the materials' thermal resistance in relation to the density, as shown in Figure 19. There is a loose negative correlation between thermal resistance and density as seen in the line of best fit, with a lower density material resulting in a higher thermal resistance. This indicates that with increasing amount of air voids, thus decreasing density, the mycelium material has an increasing ability to resist the conduction of heat flux across its thickness. The lower the value of thermal conductivity, the less heat flux is allowed from the hot surface to the cold surface; in essence a good insulator. The reverse is true for thermal resistance, the higher the value, the less heat flux is allowed and the better performing the thermal insulator. The trend observed is in line with the findings of Suleiman et al. (1999), with the cellular structure of a material resisting conduction, as voids are the dominant factor influencing heat conduction in wood materials.

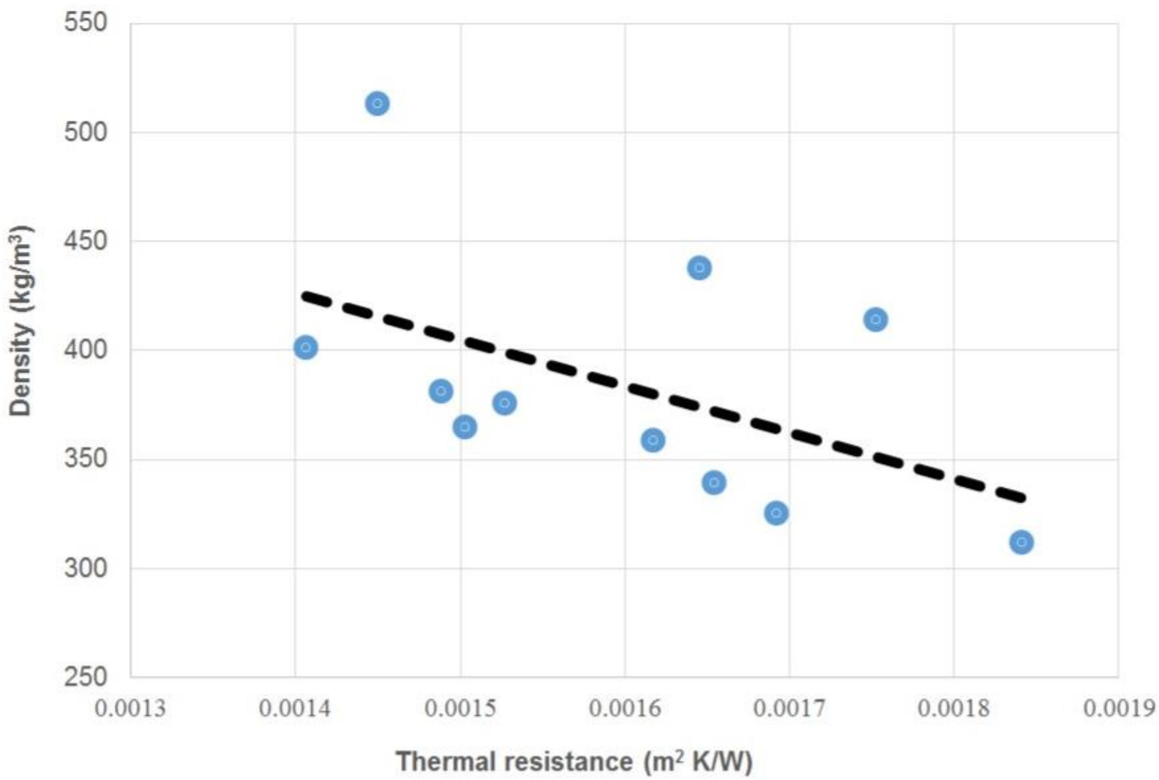

Figure 19. Plot of materials' density to thermal resistance value
for material type A, C, B1, B2, D, G, H, I , K, L and M

Thermal Characterization Conclusion

Overall, lower density mycelium materials provided thermal insulation comparable to other common natural and synthetic insulation materials, such as cork and glass fiber. For many commercial applications, such as automotive insulation, lower density materials are preferred due to supporting light-weighting efforts (ultimately reducing fuel costs). With the highest thermal resistance, R, for the lowest density, material type L was found to provide the best performance. The maximum use temperature, which is the safe temperature range within which the mycelium material would suffer no risk of smoldering or fire risk from heat sources, was found to be 300ºC (572ºF) and under, with the average maximum use temperature found to be 390 ºC (734ºF). This is higher than polymer foams such as polyurethane and phenolic foams. The findings of the efficiency of mycelium materials as an insulating material, showed overall there is a loose negative correlation between thermal resistance and density of the mycelium materials, with a lower density material resulting in a higher thermal resistance. The values for the thermal resistance and thermal conductivity ranged from 0.0014 to 0.0019 m^2 K/W and 0.053 to 0.077 W/m K respectively, which is comparable to current natural insulator materials such as balsa wood and natural cork. With increasing amount of air voids and thus decreasing density, the material has an increasing ability to resist conduction of heat flux through it. As the growth process of mycelium materials is natural, they could be tailored to provide a specific insulating R value, to fit the application the mycelium material is being designed for.

Chapter Five – Sandwich Composites: A Higher Performance Mycelium Material

Mycelium composites are a new option in the range of natural materials being investigated as alternatives to polymer-based cores for sandwich composites. Sandwich composites provide stiff and strong panels while still maintaining low density, by combining lower modulus and yield strength cores with high modulus and yield strength skins. Typically polymer foams or paper laminates are still a popular choice for sandwich composites, but both have costly manufacturing processes and use harmful chemicals in their production. Natural, less harmful materials and manufacturing processes are an increasing consideration for designers when selecting a material, and natural materials are a growing area of research to answer these needs. When mycelium composites are desiccated to a moisture content of around 5%, the result is a lightweight, insulating material, which could provide a core material for sandwich composite panels used in non-structural applications such as internal wallboards or the interior of laminate furniture.

To explore the suitability of mycelium materials for sandwich composites, the flexural characteristics were investigated with the creation of a sandwich composite comprised of mycelium composite core and carbon fiber 1-ply laminate skins. Carbon fiber was chosen as it would provide the highest stiffness for a laminate, and thus establish the upper-bounds of mycelium sandwich composites performance. The materials were made into beams of constant thickness, to make testing samples for 4-point flexural tests completed according to American Society of Testing and Materials (ASTM) C393-11e1 Standard Test Method for Core Shear Properties of Sandwich Constructions by Beam Flexure (ASTM C393, 2016). The flexural properties of mycelium matrix core sandwich composites were determined using 4-point bend tests, including flexural rigidity, core shear ultimate stress, yield strength and peak flexural stress. To investigate the feasibility of using natural skins for an entirely natural material sandwich composite, mycelium composites core with bamboo skins were created. Bamboo naturally approximates carbon fiber, being an anisotropic material with a high tensile strength and modulus in the axial direction. It also offers one of the faster growth rates of biological materials making it an excellent option in terms of manufacturing, with a fast growth rate an advantage over hard and soft woods grown for logging, helping reduce its environmental impact.

One of the purposes of these investigations is to explore the feasibility of the material for commercial uses, as part of a collaborative research program. Characterizing mycelium materials, utilizing the properties and growth parameters of fungi species, will help develop marketable controlled hybrid composite materials. Sandwich composites are often used for furniture applications, due to providing strength while being lightweight. Wood-based composites form the vast majority of low to medium cost furniture, particularly laminated wood-chip filled furniture, where compressed soft wood chip materials are formed into block shapes and laminated with polymer films to create low cost wooden tables, sideboards and other low-load bearing furniture. These materials are often commercially labeled as sustainable, defined in this context as an option which does not impact future resources (Conway & Barbier, 2013). However, the wood components are still sourced from finite resources of lumber, and can involve ecologically damaging activities. Consumers' demands for sustainable products without increased market costs continue to rise, and thus there is a real commercial need for new sustainable materials options that do not diminish limited resources. Mycelium materials could potentially offer a cost effective and sustainable option for manufacturers, as sandwich panels with a variety of laminate skins would be highly suited to several lower-load applications including furniture and decorative uses. More research is still needed into the manufacturing of these, to determine the setup and running costs, resources use of the material, and the resulting environmental impacts compared to current wood materials. Large of amounts of lignin containing biomatter is disposed of every year including wood chippings and other agricultural wastes. As a material that digests this waste to form a material made from chitin (Hiarno, 1999), mycelium-based sandwich composites could offer an attractive choice for further development to replace wood-based material for furniture.

Fabrication of Bamboo & Carbon Fiber Composites with Mycelium Cores

The naturally formed surface of mycelium materials is an uneven surface that would prevent the full adhesion of the sandwich composite skins to the mycelium materials. Thus the de-molded block of mycelium material were trimmed using a band saw to create smoothed blocks, which were then cut into the required flexural test beam samples using a rotary saw equipped with a cutting wheel. The most effective cutting tool was found to be a cutting wheel for plastic materials, chosen as toothed saw wheels were quickly blunted by the tough mycelium material. Care also had to be taken not to cut continuously, as this generated a buildup of heat that would result in the material smoldering. The frangibility of the material caused problems when trying to cut the beams to the 0.51 inch (13.0 mm) required thickness. An alternative cutting method had to be developed to prevent the rotary saw putting shear forces on the beam and causing the material to crumble, which was resolved by supporting the opposite side of the beam during cutting. The cut beam specimens were then blown free of surface debris using low pressure compressed air (15.0-20.0 psi), in preparation for the adhesion of the sandwich composite skins.

Several panels of 1-ply twill weave carbon fiber were laid up in a 0/90 degree orientation, fabricated using vacuum assisted molding, and cured for 4 hours at 176ºF (80ºC) on flat steel platens covered with mold release film and 0.197 inch (5.0 mm) polytetrafluoroethylene sheets to ensure proper mold release. Marine grade epoxy (TAP Plastics-314 Resin with TAP Plastics-102 (fast) hardener) was used as the adhesive to bond the core material and carbon fiber skin. This required the cured carbon fiber 1-ply sheets be sanded, to create a rough surface for proper bonding to the mycelium core material. After creating initial carbon fiber laminate skins to determine the most appropriate number of carbon fiber ply layers, 1-ply was determined most appropriate, as 2-ply laminates were too stiff, and would not have flexed, frustrating the aim of flexural testing.

Figure 20. Fabrication of mycelium material sandwich composites with carbon fiber laminate skins
(a) 1-ply carbon fiber skin laminate (b) Cutting of mycelium material core
(c) Unskinned mycelium composite beam samples (d) Carbon fiber skinned beam samples

Flexural Testing Methodology

Flexural tests were adapted from methodology ASTM C393-16 – Standard Test Method for Core Shear Properties of Sandwich Constructions by Beam Flexure (ASTM C393, 2016). This was selected as the most appropriate testing standard, since none specifically for the testing of mycelium materials exist yet. Testing was completed for sandwich composites with core materials of mycelium material, with skins of carbon fiber and of bamboo, chosen to provide information on the performance of sandwich composites with mycelium materials cores, with both a synthetic and a natural sandwich composite skin material. The decision to choose and test a synthetic material as a sandwich composite skin, results in higher mechanical performance, but also removes the ability to dispose of the material in a sustainable way such as through composting. The reasoning for selecting a synthetic skin for the mycelium sandwich core was to ascertain the flexural properties with the strongest and stiffest skin possible, in order to determine the upper performance bounds of the mycelium sandwich composite. Carbon fiber was selected as the synthetic sandwich composite skin due to providing excellent stiffness and strength while being low weight. Due to the difference in performance, the carbon fiber skin was likely to outperform the core material, as the synthetic woven carbon fibers have much higher strength and stiffness than a natural material. To investigate the potential of natural skin materials and determine the flexural properties of an entirely natural mycelium materials sandwich composite, bamboo was also selected as a laminate skin. Bamboo is a high strength and stiffness material in the axial directions (similar to carbon fiber), which naturally grows with these anisotropic properties. The bamboo was commercially prepared by steam treating the bamboo to form it into flat sheets, with the growth direction in the axial direction. The bamboo panels were adhered to the mycelium material core using a urethane epoxy adhesive, chosen due to having a small amount of foaming which improved the adhesion between the two uneven materials. Carbon fiber and bamboo both show strong anisotropic properties, with relatively high stiffness and strength in the axial directions compared to the transverse ones. Thus the testing samples were fabricated with the axial direction lengthwise along the beam samples, to maximize the resulting composite strength. Tests were also completed for unskinned core material, for comparison purposes. The main challenge to overcome when preparing the testing samples was the frangible nature of the mycelium composite. When cutting the samples the material was subject to shear forces and the interface between the mycelium and digested wood chips became damaged, making cutting the specimens a delicate process. The solution was to cut the blocks of mycelium composite while supporting both sides of the block to prevent the cutting forces causing shear and thus fracture. All samples were conditioned to an average 5% humidity of the core material, and all tests were carried out at standard temperature and pressure.

Flexural testing was based upon American Scientific Testing Methodology (ASTM) C393-16 Standard Test Method for Core Shear Properties of Sandwich Constructions by Beam Flexure. This testing methodology was chosen as most appropriate to determine the flexural properties of a sandwich composite with a mycelium composite foam core, covering both continuous bonding surfaces (including balsa wood and foams) and those with discontinuous bonding surfaces (including honeycomb), which the mycelium composite's properties have elements of both types. In a 3-point test the area of uniform stress is quite small and concentrated under the center loading point, however in a 4-point test, the area of uniform stress exists between the inner span loading points (typically half the outer span length), and the 4-point flexure test is common for wood and composites. Thus a 4-point bend test was selected. A 4-point bend test also best models the loading that would be experienced by the mycelium composite in potential commercial applications, such as laminated furniture or interior wall panels. An adaptation of the test specimen fixture method was necessary to accommodate the available testing equipment, inverting the fixture so the wider span was uppermost, however this had no effect on the tests or data collected. Due to the non-standard fixture a 4-point third span loading fixture installation was selected, with the dimensions shown in Figure 21, where S is the support span length of 8.0 inches (203.2 mm), L is the loading span length of 2.5 inches (63.5 mm), and P the loading applied, as according to ASTM C393. The support span must be sufficiently short so that transverse shear forces are produced at applied forces low enough, and the transverse shear forces are being carried by the facings, giving falsely high apparent core strength. To prevent this, the span-to-thickness ratio was constrained to 14, except in the case of the bamboo samples which were fabricated with a ratio of 10, due to manufacturing constraints of the samples.

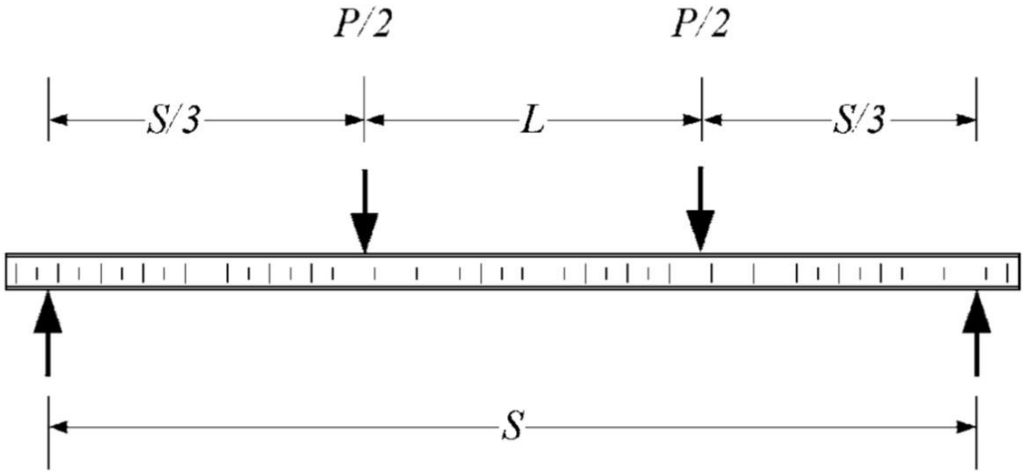

Figure 21. ASTM C393 4-point third span testing fixture (ASTM C393, 2016)

Tests were conducted at a deflection rate of 0.236 inches/minute (6mm/minute) until failure occurred for all tests. As moisture level affects mycelium material properties, all samples were standardized to a moisture content of 5.0% core humidity, measured with a digital moisture meter and verified immediately prior to testing. Five tests were conducted for each type of beam sample, the results averaged, and the force deflection results used to calculate the core shear ultimate stresses according to ASTM C393 as shown in Equation (22) (ASTM C393, 2016), where F_{ult} is the core shear ultimate strength in MPa, P_{max} is the maximum force prior to failure in N, d is the sandwich thickness in mm, c is the core thickness in mm, and b is the sandwich width in mm.

$$F_{ult} = \frac{P_{max}}{(d + c)\, b} \qquad (22)$$

ASTM methodology D790-17 (ASTM D790, 2017) was used to calculate the flexural stress and flexural strain based on homogenous material using beam flexure theory, and the modulus calculated from the resulting stress strain plots. During the tests a Zoom High Definition video recorder was used to capture the tests, and images captured from the video, as seen in Figure 22, were used to analyze the testing specimens' failure modes.

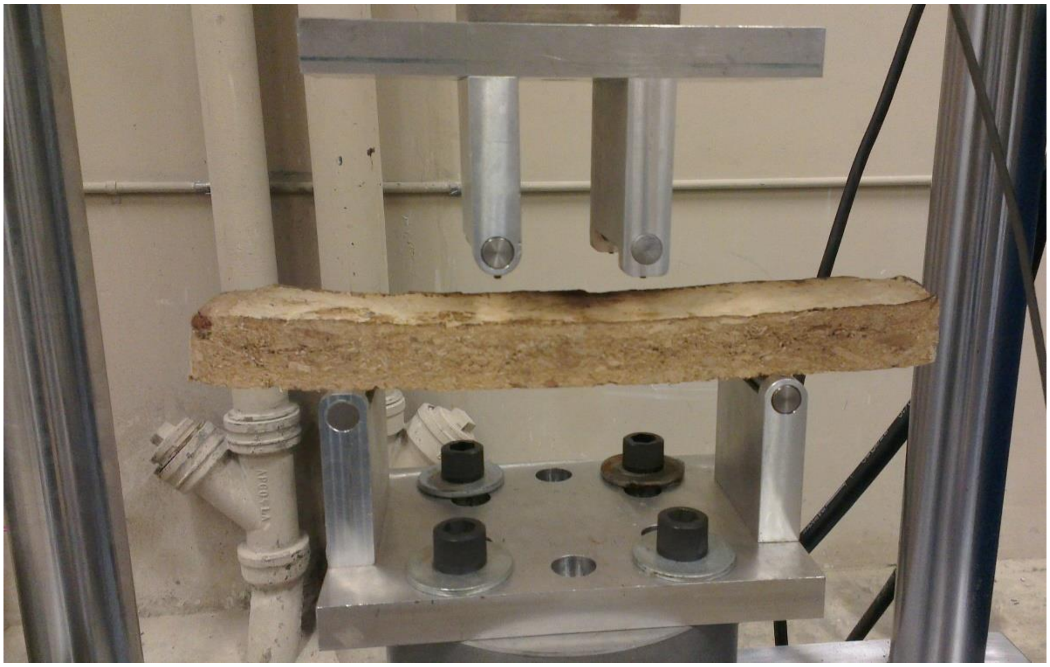

Figure 22. ASTM C393 4-point third span testing fixture setup with unskinned core beam sample

Core Shear Ultimate Strength: Sandwich Composites Double Performance

After inspecting the testing video recordings, the carbon fiber skinned samples failed in mode D, skin to core delamination. The cause was identified as brittle failure of the epoxy resin used to adhere the facing to the core material, causing the facing skins to peel away from the core material and reducing the failure strength of the sandwich. The core material in the carbon fiber skinned sandwich beam sample also showed failure with crack propagation across the core material at 45 degrees to the horizontal, suggesting a brittle failure mode which supported the observations of the core material's frangibility during sample manufacture. The bamboo skinned samples failed in a mixed mode, first characterized by mode S, transverse shear at approximately halfway through the thickness of the sample, and with mode D, skin to core delamination. The skin to core adhesion performed better than the carbon fiber skinned samples, with the top skin of the bamboo beam samples staying intact, and only the bottom skin delaminating. From this the urethane adhesive was determined as a better adhesive option, due its higher toughness and ability to maintain cohesion under bending moments. The slight foaming of the urethane adhesive may also have contributed to better bonding of uneven surfaces and thus performance. The mycelium composite core only beam samples with no skin facings failed though mode S, transverse shear causing cracks to propagate across the core material at 45 degrees to the horizontal, indicating a brittle failure mode. This indicated brittle failure of the material, which is as expected for the material which has frangible properties.

The core shear ultimate strength was calculated from the maximum force prior to failure for each type of beam sample, according to ASTM C393 method described in Equation (22). The results are summarized in Table 6. The unskinned achieved core shear ultimate strength of 36.2 kPa, the mycelium composite core with carbon fiber skins achieved a core shear ultimate strength of 76.6 kPa, and bamboo skinned beam samples achieved 63.3 kPa. These results indicate the addition of skin facings increase the strength of the sandwich composite. Carbon fiber laminate skins provided the best performance improvement, increasing the core shear ultimate strength by 112%, and the bamboo skins provided an improvement of 75%, in comparison to unskinned beam samples.

Beam sample type	**Maximum force prior to failure (N)**	**Core shear ultimate strength (kPa)**
Mycelium composite core only	43.6	36.2
Mycelium composite core with carbon fiber skins	95.6	76.6
Mycelium composite core with bamboo skins	209.7	63.3

Table 6. Core shear ultimate strength by beam sample type

Stress strain plots were created from the testing data of the flexural tests, as shown in Figure 23, 24, and 25. All the beam samples showed elastic deflection up to their yield strength, followed by plastic deformation and finally brittle failure. After failure samples continued to deflect, producing small forces, until tests were manually stopped. The carbon fiber skinned beam samples, and core only beam samples showed consistent load deflection plots, yield strengths and failure strengths; however, the bamboo skinned beam samples showed more scattering of data points. Some of this was identified as due to the natural variability of the bamboo material properties, which is inherent to natural materials, and some from a noise problem from the hydraulic testing machinery which was corrected during the testing. Despite delamination, the samples stayed largely intact during deflection, retaining wholeness in spite of cracking and eventual failure. This has positive implications for potential sandwich composites applications, as during failure, the material will stay cohesive. This is a useful attribute for materials used in non-structural applications, where failure is aesthetically important as opposed to critical for safety. For example, materials used for furniture need to retain their appearance of integrity to be considered pristine, even if this does not impact their functional properties. Thus, mycelium materials composites which will remain whole even when damaged, suggests they would be useful for furniture applications.

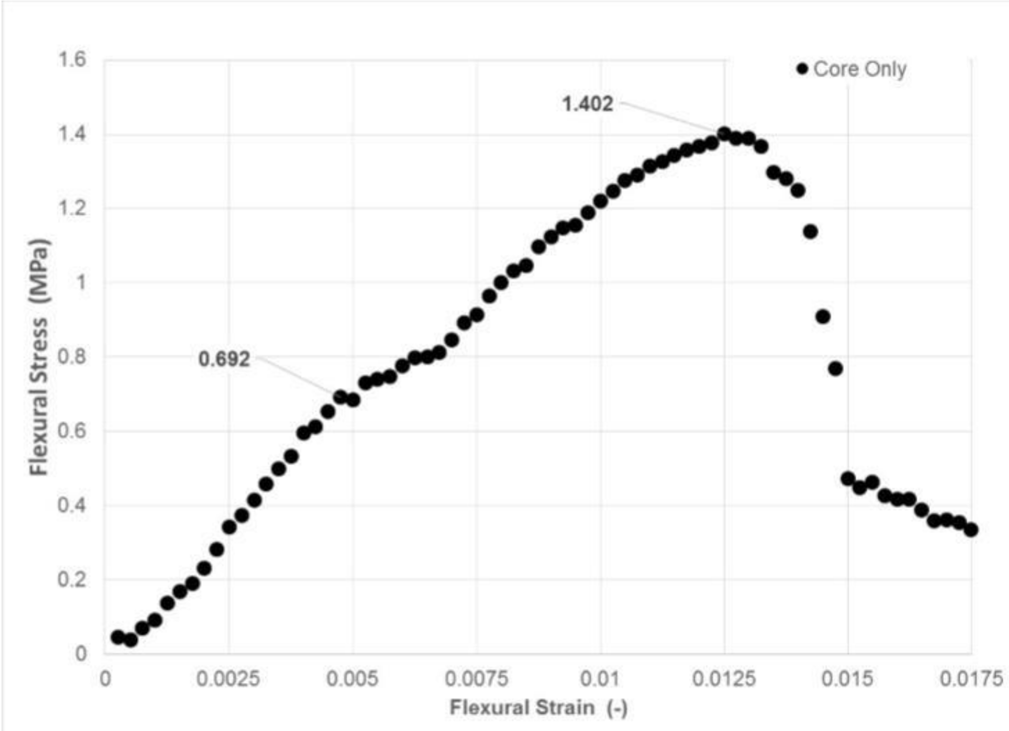

Figure 23. Stress strain plot of mycelium material unskinned core flexural test

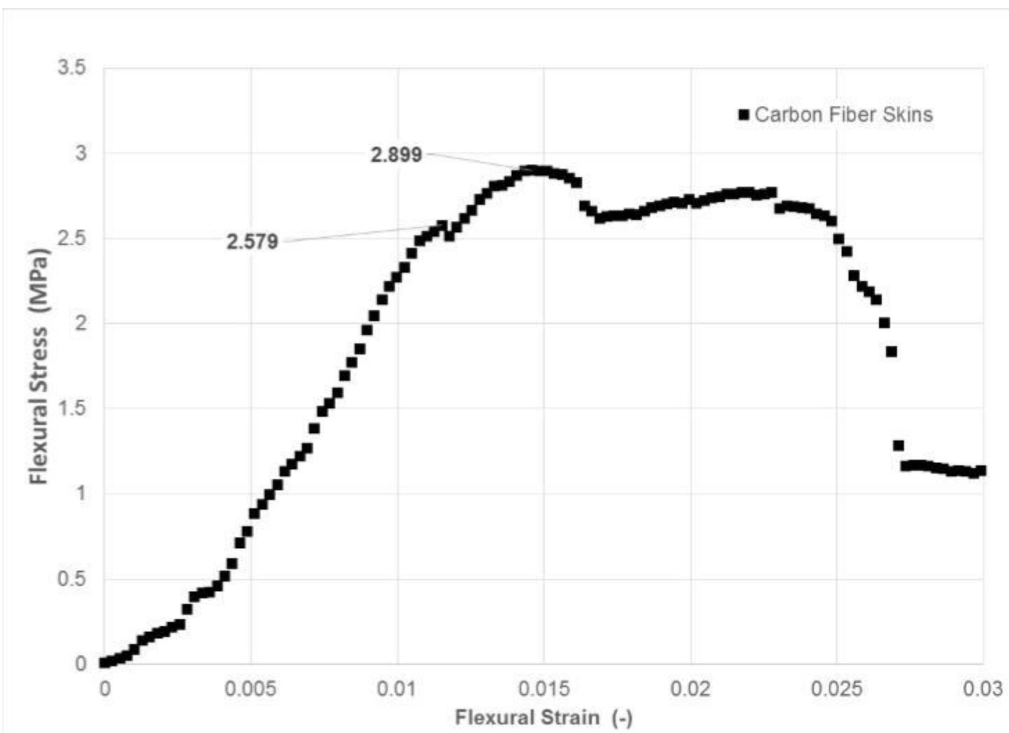

Figure 24. Stress strain plot of mycelium material core and carbon fiber skins flexural test

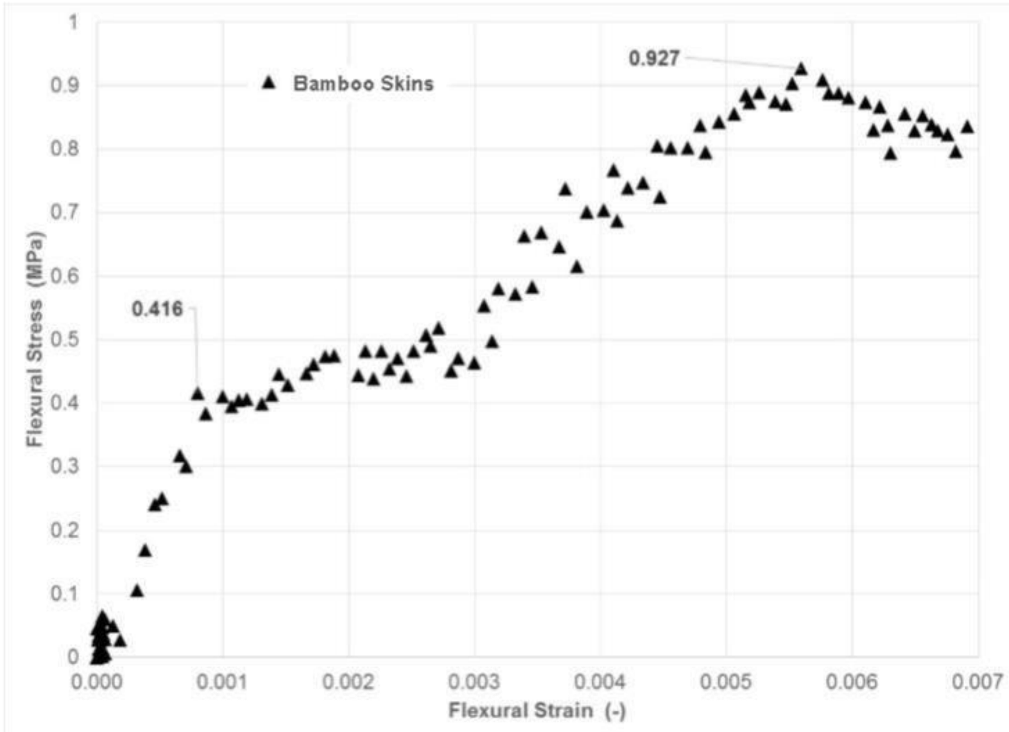

Figure 25. Stress strain plot of mycelium material core and bamboo skins flexural test

The unskinned core material developed a flexural modulus of 168 MPa, as seen in Figure 23, until the flexural yield strength of 0.7 MPa. The yield point was definite but not characterized by a drastic drop in forces, with the material undergoing mainly plastic deformation until the peak flexural stress of 1.4 MPa, followed by a steep decrease in the flexural strength, before failure and the subsequent linear deformation from the loading actuator while the sample was failed and broken.

The results of the carbon fiber skinned beam samples, as seen in Figure 24, showed the modulus was increased by 76% to 296 MPa by adding carbon fiber facings. This is consistent with the high modulus properties of carbon fiber material reinforcing the lower modulus of the mycelium composite. The flexural yield strength increased by 271% to 2.6 MPa, and the peak flexural strength also increased by 107% to 2.9 MPa, indicating the carbon fiber facings increase the strength and modulus of the beam samples due to reinforcing the weaker core material. This suggests the mycelium material would be an ideal low density and lower strength mycelium core material to be reinforced by synthetic skins, which increase its strength and stiffness.

The bamboo skinned sandwich composite, shown in Figure 25, demonstrated a stiffer flexural modulus of 645 MPa, exceeding the modulus of the carbon fiber skinned composites by 118%. This was due to the thicker facing skins aiding greater flexural strength, better adhesion of the core to the facing skins, and the properties of the axially orientated bamboo fibers reducing deflection during loading. The yield strength of the bamboo sandwich composites was 0.4 MPa and the peak strength was 0.9 MPa. They did not achieve the same peak strengths as the unskinned and carbon fiber skinned sandwich composites, likely due to the bamboo beams low span-to-thickness ratio of 10. This resulted in transverse shear forces being transferred to the facings, and thus not using the full strength of the combined core and facings, resulting in the lower peak strength.

In summary, sandwich composite beams of unskinned core mycelium material achieved a core shear ultimate stress of 36.2 kPa, with a flexural modulus of 168 MPa, a flexural yield strength of 0.7 MPa, and peak flexural strength of 1.4 MPa. The addition of carbon fiber skins much improved the flexural properties, achieving 76.6 kPa core shear ultimate stress, a 76% improved flexural modulus of 296 MPa, 271% improved flexural yield strength of 2.6 MPa, and the peak flexural strength also increased by 107% to 2.9 MPa. This indicates synthetic skins offer the most reinforcement and improvement of the mycelium composite properties. The natural sandwich composite of mycelium composite core material and bamboo laminate skins achieved slightly lower core shear ultimate stress of 63.3 kPa, flexural yield strength of 0.5 MPa, peak flexural strength of 0.9 MPa, due to a span-to-depth ratio of only 10 (the ideal being 14). The natural sandwich composite did achieve a much improved flexural modulus of 645 MPa, exceeding the modulus of the carbon fiber skinned composites by 118% due to the thicker facing skins aiding greater flexural strength, better adhesion of the core to the facing skins, and the properties of the axially orientated bamboo fibers adding to the combined strength of the material.

The main issue encountered was compliance with the core thickness to length requirements to minimize core shear within the beam specimen. The blocks of mycelium composite grow to a maximum mold dimension of 12.0 inches (305.0 mm) by 5.0 inches (127.0 mm) by 1.25 inches (32.0 mm), limiting the maximum beam span possible. This in turn constrained the minimum core thickness to the range of 10-14. A preliminary test with rubber pads in place to ensure no point loading effects of the beam specimens, which could crush the material rather than induce deflection, showed the pads caused slippage of the specimens in the testing fixture resulting in noise in the readings. To resolve this, subsequent tests were completed without pads. This may have increased the point loading effects which may have in turn increased the likelihood of failure by de-bonding of the face sheets. Other challenges to be overcome also include the adhesive bonding strength and flexibility of the bonding material used to prevent facing skin delamination. Given the benefits of creating an entirely natural sandwich composite, investigation of natural adhesives such as rabbit skin glue would be a useful next step to investigate.

Despite the span-to-thickness ratio issue, the results showed mycelium materials create an entirely natural composite, composed of materials which are sustainable, being grown from waste. It also showed mycelium materials can deliver a strength of 1 MPa and a stiffness of 645 MPa, which is comparable to polymeric foams (Ashby & Johnson, 2013). These merit the material as having the potential to be developed further. An entirely natural sandwich composite, composed of mycelium materials and natural laminates such as bamboo, can offer unique advantages of sustainability, due to being compostable. Materials which can be grown from waste agricultural materials, such as husks from food crops, and that can be ecologically disposed of while also not requiring the use of petrochemicals, show promise to be investigated for applications such as interior wall panels and non-structural uses of panels. While some challenges need to be overcome, including the adhesive bonding strength needed to prevent facing skin delamination, sandwich mycelium cored composites show good potential replacement material.

Sandwich Composites Characterization Conclusions

The core shear ultimate strength of the unskinned core mycelium material was identified by finding a core shear ultimate stress to be 36.2 kPa, which was improved to 63.3 kPa with bamboo skins and 76.6 kPa with carbon fiber skins. This shows mycelium sandwich composites would be suited to low-load applications in the 60-76 kPa range. As the both the carbon fiber skinned sandwich samples and the bamboo skinned sandwich samples both failed in mode D, skin to core delamination (with bamboo also failing in a mixed mode), reducing delamination would be a key area of focus to improve the materials properties. While the urethane adhesive does not allow for a totally natural composite, which impacts disposal options, it was found to be a better adhesive option, due its higher toughness and ability to maintain cohesion under bending moments.

Overall, due to offering a low cost, low-density cellular core material grown from sustainable and renewable resources, mycelium materials have great potential as a core material for sustainable sandwich composites. Both the synthetic skins and bamboo skinned sandwich composites show potential to be developed further, also adding the advantage of increased moisture ingress resistance of the material. Sustainable materials continue to grow in demand, and composite materials which can be grown from waste agricultural materials, ecologically disposed of by composting, and which do not require the use of oil-based chemicals in their fabrication, show promise to be investigated for applications such as interior wall panels and non-structural uses of interior paneling.

Section One Summary: A Replacement for Polymer Foams & Sandwich Composites

The novelty of mycelium-based materials means there was a need for empirically determined material properties, which turn required an understanding of the structure and resulting physical properties of the mycelium-based material. Optical microscopy identified mycelium growth as a white colored mass of hyphae, concentrated around the feedstock chips. Scanning electron microscopy identified the random, tangled nature of the hyphal growth, showing a structure closer to a synthetic fiber random-mat structure, than the theorized cubic open-walled cell structure. This helped lend insight into the insulation properties of mycelium materials, as the air spaces in-between the hyphae help prevent effective heat transfer through the material.

Using asymptotic homogenization, a model was developed to explain the deformation of the cellular structure under load, determining the material's Young's Modulus of 0.96 MPa, and experimentally determined as 1.3 MPa. Under tensile loading the material showed the expected pattern for an elastic-plastic material, with a linear-elastic region up to a 0.2% offset yield point of 15 kPa, a negligible plateau region, and finally plastic deformation until brittle fracture at 176 kPa. The relatively low Ultimate Tensile Strength of 176 kPa reflected the brittle and frangible nature of the material. The material stiffness in compression was calculated as 1.0 MPa, and the compressive yield found as 47.5 kPa. The ultimate compressive strength of 490 kPa was around three times the UTS. This makes the material comparable to polymer foams, such as starch-based foam, and polystyrene foams.

To improve the mechanical performance, sandwich composites were created with carbon fiber and bamboo laminate skins with a core beam of unskinned core mycelium material. The results are summarized in Table 7. The addition of carbon fiber skins much improved the flexural properties, achieving a +76% improved flexural modulus, a +271% improved flexural yield strength, and 107% peak flexural strength. This indicated synthetic skins offer the most reinforcement and improvement of the mycelium composite properties.

The natural sandwich composite of mycelium composite core material and bamboo laminate skins achieved slightly lower core shear ultimate stress, but also achieved a much improved flexural modulus, exceeding the modulus of the carbon fiber skinned composites by 118% due to the thicker facing skins aiding greater flexural strength, better adhesion of the core to the facing skins, and the properties of the axially orientated bamboo fibers.

	Core shear ultimate stress	**Flexural modulus**	**Flexural yield strength**	**Peak flexural strength**
Unskinned mycelium core	36.2 kPa	168 MPa	0.7 MPa	1.4 MPa.
With carbon fiber skins	**296 MPa**	296 MPa	**2.6 MPa**	**2.9 MPa**
With bamboo skins	63.3 kPa	**645 MPa**	0.5 MPa	0.9 MPa

Table 7. Summary of mycelium materials flexural properties

The maximum use temperature, defined as the safe temperature range within which the mycelium material would suffer no risk of smoldering or fire risk from heat sources, was found to be 300ºC (572ºF) and under, with the average maximum use temperature found to be 390 ºC (734ºF). This is higher than polymer foams such as polyurethane and phenolic foams. The values for the thermal resistance and thermal conductivity ranged from 0.0014 to 0.0019 m^2 K/W and 0.053 to 0.077 W/m K respectively, which is comparable to current natural insulator materials such as balsa wood and natural cork.

Overall, the cellular structure of mycelium and mechanical and thermal properties means it can be considered comparable to polyurethane and phenolic foams, and it suited to being a core material for sandwich material. Barriers to development and adoption of such materials in place of polymeric options, include the potentially higher costs and development efforts needed due to the novelty of the material. Further development is needed to improve the strength of mycelium materials, and to explore the range of properties that can be achieved with sandwich composites.

SECTION TWO: GROWING, DENATURING & FORMING MYCELIUM MATERIALS

A key part of understanding mycelium materials behavior and thus suitability for applications, is the consideration of the fabrication and manufacturing processes required to form useful materials. The following investigation into the subtractive and thermal cutting of the material explores how the material performs in forming basic and detailed features, including the use of pyrography to form surface decorations. This section also aims to answer the question many designers have when discovering the material; how it degrades in exterior and interior environments, along with briefly exploring the fabrication, denaturing and post-processing of mycelium materials.

Chapter Six – Mechanical Subtractive Manufacturing

Once inert, mycelium materials undergo manufacturing processes to form a variety of products, including traditional machining such as drilling, shaping, and finishing processes. Part of the characteristics of mycelium materials is their response to manufacturing activities, including commonly used fabrication methods such as material removal processes. Understanding mycelium materials response to these processes is important for manufacturers and developers (Jiang et al., 2019), and as each type of material varies greatly in its behavior under cutting tools. An empirical method has been used to determine their suitability for use in different applications, including those on which the resulting character of the machined surface is important, by determining their performance in common subtractive machining methods. This includes applications where mycelium material is exposed, such as in artisan products, and can also include products where the mycelium material is interior, such as natural sandwich composites. The suitability and resulting finish that traditional manufacturing processes for mycelium materials is a new area of research. As applications for mycelium materials increase, there is an emerging need for understanding the material's behavior during fabrication and manufacture. Determining the response of mycelium materials to commonly used manufacturing methods is useful to both commercial and artisan users, as the results can be used to inform the development of applications that utilize the material. Common manufacturing includes cutting and abrasive processes to form the material from its original in mold shape.

Figure 26. Image of cut mycelium material samples

Testing was carried out to determine the effect and results of various manufacturing processes on the mycelium material, according to the testing methodologies of American Society for Testing and Materials. Experimental testing also included assessment and discussion of the results for various cutting processes including milling, drilling, and sanding. Due to the novelty of the field of mycelium materials, no specifically designed testing standard existed for wood-based mycelium composites, so the testing methodology was adapted from American Society for Testing and Materials D1666 – 17 Standard Test Methods for Conducting Machining Tests of Wood and Wood-Base Panel Materials (superseding D1666-11) (ASTM 1666). This was selected as being most appropriate due to the wooden feedstock and composition of the material, and wood panel-like composition with a differing mycelium material surface composition, to the interior mycelium material.

Since the mycelium materials are grown in smaller geometries than the wood products referred to in the ASTM D1666 standard, the test sample sizes were reduced to accommodate the available materials. Five test specimens were visually examined for specified defects after each run. As according to ASTM D1666, each test specimen was inspected to classify characteristics by visual examination based on five grades or groups:

- Grade 1, excellent
- Grade 2, good
- Grade 3, fair
- Grade 4, poor
- Grade 5, very poor.

Visual inspection & classification of defects was carried out, including:

- Chip marks, defined by ASTM D1666 as shallow dents in the surface caused by shavings that have clung to the cutting blades instead of passing off in the exhaust as intended
- Fuzzy grain, defined by ASTM D1666 as small particles or groups of fibers that did not sever clearly in machining but stand up above the general level of the surface.
- Breakouts; defined as frangible areas disturbed or broken from the stock material
- Scratches; defined as shallow or deep grooves that damage the material's surface

The qualitative nature of the grading scale was found to be adequate for determining the quality of the finishes, with the test specimens showing good consistency, and showed mycelium materials can be graded in a similar way to wood materials. Overall the mycelium materials performed similar to wooden materials, and offered additional forming and molding options during fabrication of the naturally grown, fungi-based mycelium material. Since the test results represent subjective and comparative classification characteristics from visual examination, no statement is made about either precision or bias of the results.

Milling

The test equipment for the milling test was selected to represent typical fabrication equipment used in the production of small to medium volumes of mycelium material based products. A handheld routing power tool was selected with a spindle speed of ~20,000 rpm, and feed rate 39.4 inches (1000 mm) per minute. A single groove of 1⁄4 inch (6.0 mm) was cut inboard from and parallel to the preliminary cut, with a groove depth of 1⁄4 inch (6.0 mm). The groove was cut as a straight line for the full length of the specimen. Following the test, each of the five test specimens was examined, inspecting the groove for breakouts, sharp corners, chipping, fuzzy edges, and the general smoothness of cut, evaluated using the scale described above.

Prior to milling, the five test specimens were identified as Grade 2, with a good surface finish and a napped texture, with some of the surface morphology color variation that is typically seen in natural materials. The five tests were carried out using the above methodology, and the test specimens examined. The test surfaces were identified as Grade 1, excellent. The specimens' grooves were inspected for breakouts, sharp corners, chipping, fuzzy edges, and general smoothness of cut. The grooves milled into the test specimens showed evidence of no evidence of breakouts, sharp corners, chipping, of fuzzy edges, and developed an overall a medium smoothness of cut. The groove showed an excellent cut, with a precise and well-formed profile 90 degree angle of the milled groove, indicating milled cuts can be made with a high degree of accuracy and tight tolerances.

Figure 27. Milling test specimen showing profile of milled grooves.

The tests also led to noting that the composition of the material, including its density and type based on the species, will have a strong impact on the efficacy of the manufacturing method. Denser, harder materials are likely to have better cutting performance, whereas the less dense, more frangible materials will perform worse, due to cutting processes requiring a well-consolidated material to plastically form chips, also reducing brittle fractures.

Drilling

The test equipment for the drilling process was also selected to represent typical fabrication equipment used for mycelium materials. A single spindle electric drill machine equipped with power feed was selected, and 5 holes bored into the test specimens. The drill bit selected was a 1/8 inch (9.5 mm) twist drill with a 120º point. The spindle speed was set at ~3500 rpm, to drill holes to a depth of 1 inch (25.4 mm). Following each test, the test specimens were visually examined. The holes were inspected and graded, and inspected for chipping, fuzzing, thickening of the edges, and general smoothness of cut, using the visual inspection scale described above.

Prior to testing the test specimens' surfaces were identified as Grade 2, showing a smooth, good surface with some texture and surface morphology variation typical of natural materials. The 5 tests were carried out according to the above methodology, and visually inspected.

All 5 tests showed after drilling the surface surrounding the drilled hole was determined to be Grade 5, very poor, with substantial chipping and fuzzing. This was due to the material suffering brittle fractures during the cutting process, around the entry point into the material. This is potentially due the differing properties of the surface and interior material, and potentially the spindle speed and cutting head angle. To fully determine how to improve the properties, further testing should be completed on various spindles speeds and cutting head angle. This would help determine if varying the parameters can produce less chipping and fuzzing. As the milling process was removed the material through a chip-forming process, the spindle speed is the most likely parameter to focus on for better drilling performance. The drilling slower spindle speed of ~3500 rpm, compared to the milling spindle speed ~20,000 rpm which produced good cutting performance, indicates the poor drilling performance may be improved by increasing the spindle speed. Visual inspection on the interior of the drill holes found the interior surface finish to be Grade 3, showing fair surface smoothness with some fuzzing. The interior material is more consistent, lacking the more frangible napped surface mycelium materials naturally form, and this is likely why the surface finish of the holes is better than the area surrounding the drill entry point.

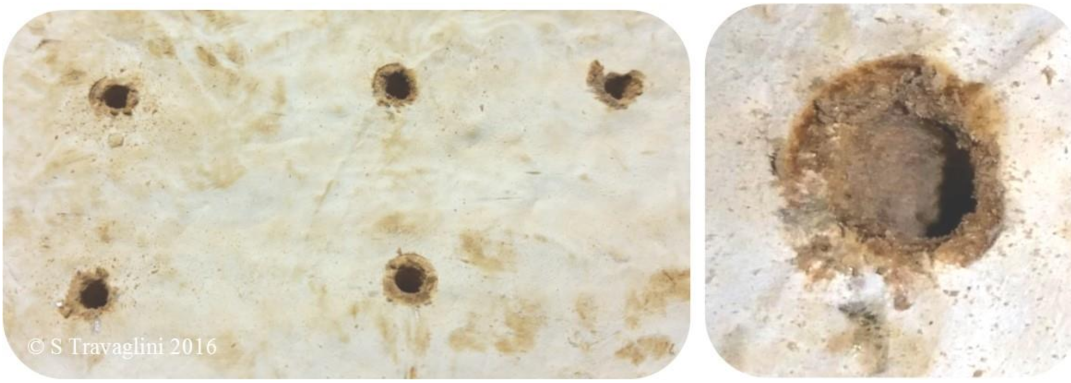

Figure 28. Drilling test specimen showing five test holes (left), and drilled hole close-up (right).

<u>Sanding</u>

The sanding test equipment was selected to represent a commonly used fabrication equipment, a single headed powered belt sander hand tool. The sander was equipped with an aluminum oxide sanding belt of 80 and 120 grit, as according to ASTM D1666. The feed rate was approximately 20 ft/min (6.1 m/min), and the depth of cut was 1.5 mm. Following each test the 5 specimens were examined for the surface finish and defects such as scratching and fuzzing, using the visual inspection scale described above. The test specimen surfaces prior to the tests were identified as Grade 2, a good surface finish with an overall smooth, and with natural texture and surface morphology variation. Following sanding tests with the 80 grit belt, a Grade 4 surface finish was developed with some fuzzing and variation in smoothness, as seen in Figure 29. The test specimens sanded with a 120 grit belt were inspected, and found to have a Grade 3 surface finish with a small amount of fuzzing, as shown in Figure 30.

Figure 29. Sanding test specimen surface with 80 grit, showing surface Grade 2 - Good surface finish before (left) and Grade 4 - Poor surface finish after (right).

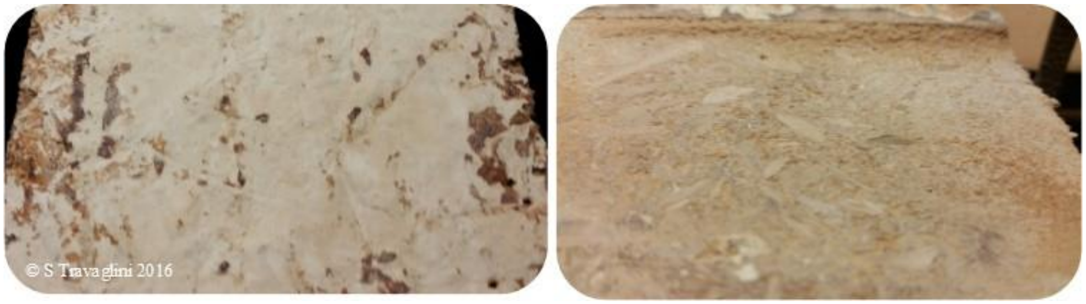

Figure 30. Sanding test specimen surface with 120 grit, showing Grade 2 - Good surface finish before (left), and Grade 3 - Fair surface finish after (right).

The finer 120 grit abrasive process resulted in a finer surface finish in comparison to the 80 grit test specimens, with the mycelium material behaving similar to wood-based materials. This fitted with the composition of the mycelium materials feedstock of cellulose-rich sawdust. Both sets of tests offered a smooth, consistent, planar surface, making the materials surface suitable to be coated with paints or natural sealants.

Traditional Manufacturing Process Conclusions

Drilling, milling and sanding performance showed mycelium materials can achieve a fair surface finish with traditional manufacturing processes. The frangible nature of the material does result in defects including chipping and fuzzing. Milling achieved the best surface finish of the three subtractive cutting methods, with no chipping or fuzzing, and achieving a precise cut profile featuring well-defined 90 degree angles. This leads to the conclusion that mycelium materials are suited to fabrication by CNC milling, offering a precise cut and good surface finish. Drilling resulted in a fair surface finish for the bore of the drilled hole, but a very poor finish for the top of the mycelium material surrounding the holes, suffering chipping and uneven material removal. Based on this, it is concluded drilling is a suitable cutting method for boring and pre-drilling, for example for fixings, but requires counter-sinking or other refinement post-drilling if a good surface finish is required. Sanding developed a fair surface finish, with finer surface finishes produced with higher grit grades of abrasive. Inspection of the chips formed during the cutting processes showed the chipping and fuzzing of the material is partly due to small pieces of the material breaking off during the cutting processes. Chips broken from the material also tended to occur around larger chunks of un-digested feedstock, potentially indicating a less strong bond between the feedstock chips, and the resulting mycelium material itself. The materials performed in a similar way to wooden materials which fits with the mycelium materials' feedstock being wood-based, with some undigested feedstock remaining in the mycelium material. All of the methods were deemed suitable manufacturing methods for material removal, with milling offering the best surface finish and precision of cut. While the visual grading scale adapted from American Society for Testing and Materials D1666 – 17 (Standard Test Methods for Conducting Machining Tests of Wood and Wood-Base Panel Materials) was satisfactory in terms of identifying the surface finish, the qualitative method is open to interpretation. The aim of the testing was to determine the suitability of cutting methods to inform designers & fellow engineers considering using the material for various applications, so expressing the surface finish in general terms is adequate for these needs. Improvements to the method could be the inclusion of surface roughness metrology, expressing the Roughness Average (RA) of the materials. However, the RA scale may be too precise for the macro-scale of the mycelium materials surfaces, so another option could be comparison of geometric tolerances of the material thickness at various locations on the test specimens.

Further exploration into the effect of the type & density of the material would provide more information on suitable manufacturing methods for a wider range of materials. Exploring the difference in material properties between the surface and interior of the mycelium materials would also provide more information for designers, when choosing suitable materials & their manufacturing options for various applications. For example, mycelium materials used in sandwich composite applications may need sanding to form surfaces for good composite skin adhesion, whereas materials for insulation or automotive parts would be molded so sanding would not be needed. In each case, the choice of fungi species and materials type would affect the choice of manufacturing method and vice versa. Further investigation of the impact on each manufacturing process, from the differing material properties of the surface and interior, different spindle speed, and cutting head angle, would also provide more information on the effect on surface finish from varying the process parameters.

Chapter Seven – Non-traditional Subtractive Manufacturing

While mechanical cutting processes are suited for bulk shaping processes, for example forming dimensionally precise bricks from molded blocks of mycelium materials, finer features are harder to achieve due to the frangible nature of the mycelium materials. While milling offers a good surface finish, cutting processes result in chipping and fuzzing of the material. The chipping and fuzzing is due to the frangible nature of the material, which may occur with all mechanical cutting processes. This makes forming making small features, around a quarter inch, difficult to achieve.

Non-traditional processes that don't involve mechanical cutting could provide alternative shaping methods, providing designers and developers with more options for fabrication processes, particularly if the application requires creating small features. Evaluating shaping and cutting processes as alternatives to mechanical processes will help provide the information needed for fabrication & applications in this emergent field, by informing developers & designers of the most suitable manufacturing options for mycelium materials. Five common non-traditional manufacturing techniques, as described by Benedict (2017), were considered as alternatives:

- Water jet machining
- Ultrasonic machining
- Electrochemical discharge machining
- Electrical discharge machining
- Thermal cutting

An initial evaluation of each of the methods listed above was carried out, to select the most suitable method for further empirical testing. This was based on the suitability of the materials removal method for mycelium materials, and availability of equipment for each method.

Water jet cutting, a high pressure stream of water which removes material through abrasion, offers the ability to cut through very hard and dense materials. However, the method has a high potential for water damage and water ingress into the material. Mycelium materials are low density, and are desiccated to around 5% moisture content during their fabrication processes. Desiccation is used to ensure that the material does not degrade, or suffer reduced material and mechanical properties, due to an excess moisture level. Any process that introduces moisture is not a viable method to cut the material, and thus water jet cutting was determined to be unsuitable and rejected for further evaluation or testing.

Electrochemical discharge machining and electrical discharge machining use electrostatic thermal reactions to remove material particle by particle. Both require the material to be electrically conductive, to create the charge difference between the material and removal tool which leads to the removal of the material particle. Mycelium materials are non-electrically conductive, based on a basic test using a multi-meter to determine if electrical current could pass through the material, which showed the multi-meter did not register any electrical conduction through the mycelium materials. Due to this, both electrochemical and electrical discharge machining were determined to be unsuitable for further evaluation or testing.

Ultrasonic cutting still involves a cutting blade, along with ultra-high frequency vibration of the cutting blade, which provides good performance when cutting brittle materials (Feucht et al, 2014). Ultrasonic cutting is used in industry for a wide variety of materials, including textiles, concreate, and edible foods such as bread. Bread can be described as having a cellular structure and low tensile strength and fracture toughness, which is similar to mycelium materials. This, coupled with the good performance when cutting brittle materials, makes ultrasonic a good potential cutting method. Ultrasonic cutting is not a widely used process in traditional machine shops, and no ultrasonic cutting machines were available for testing or further evaluation. However, they have good potential for evaluation in future work.

Laser cutting uses a focused beam of light to heat and vaporize a material in small sections. This is a thermal cutting process, which incinerates the material without any mechanical interaction with the material. This could reduce or eliminate the fuzzing and chipping associated to mechanical cutting processes, but could potentially cause excessive burning or charring of the material. As a 'non-traditional' process, thermal cutting is known for creating precise and detailed cut patterns in a variety of materials, and also for producing decorative surface patterns through pyrography. Laser cutting removes material by transferring energy using a focused beam of light on a material, resulting in the localized temperature in a small area rising above the material's melting temperature, and melting or vaporizing the material (Caristan, 2004). The behavior of mycelium materials during combustion is similar to thermoset polymers, likely due to a carbon-based structure with permanent cross-links, except with organic protein-based chitin instead of the double carbon bonds formed in polymers. During combustion the chitinous material degrades into carbon products, such as CO_2 and char (Horrocks & Price, 2008). Thus the energy required to cut the mycelium material, must exceed the energy required to combust the chitinous structure. This means that surfaces exposed to laser cutting will exhibit signs of thermal degradation, including charring, which may result in excessive damage to the material during laser cutting.

Laser cutting was selected to be further evaluated, based on process suitability and equipment availability. To determine the suitability of laser cutting, 3 tests were designed to evaluate the performance of; bulk cutting, small feature cutting, and pyrography. These were selected as typical fabrication needs for consumer applications, such as forming dimensional blocks from bulk volumes of the mycelium material, or the creation of logos and decorative features as artistic and aesthetic finishing processes.

Establishing Laser Cutting Parameters

No specifically applicable testing standard existed for laser cutting mycelium materials, so testing methodologies were developed for the laser cutting testing as informed by ISO 9013:2017 Thermal cutting - Classification of thermal cuts (ISO 9013, 2017). The required parameters, such as power intensity and speed of cut, first had to be experimentally determined, due to this research being the first instance of laser cutting technologies being tested on mycelium materials.

The laser testing equipment was a Universal Laser System PLS6MW machine, with a CO_2 laser with a cutting power of 60 Watts, with a beam width of 4.17×10^{-4} inches (10.6μm), and maximum linear speed of 70 inches/second (1.78 m/s) with 1000 pulses/inch (25.4mm). The required power and speed needed to penetrate and remove the material will be determined by creating a range of cutting speeds, all with 100% power of 60 Watts, in a series of short linear cuts of 1 inch (25.4 mm) each, according to the following parameters detailed in Table 8 below. The laser cutting power was set at 100% (60 Watts), to ensure the laser beam fully penetrated the material, and the speed used as the dependent factor in determining the best cutting parameters.

Cutting Section	**Cutting Speed (%)**	**Cutting Speed (inches/second)**	**Power (Watts)**	**Pulses Per Inch**
A	5	0.70	60	1000
B	4	1.40	60	1000
C	3	2.10	60	1000
D	2	2.80	60	1000
E	1	3.50	60	1000

Table 8. Bulk laser cutting testing parameters

The testing sample was a thin flat slab of the mycelium material, selected to include the tougher chitin-rich surface layers, while ensuring an achievable cutting volume of the interior material with a thickness of 0.12 inches (5.0 mm). The test cuts were evaluated in terms of depth of cut penetration, dimensional accuracy and precision of cut, and degree of surface damage such as charring on a qualitative scale of 1 to 5, chosen to meaningfully describe the impact of the cutting operation on the aesthetics of the material:

1 – No damage
2 – Visible damage
3 – Mild damage
4 – Significant damage
5 – Extensive damage

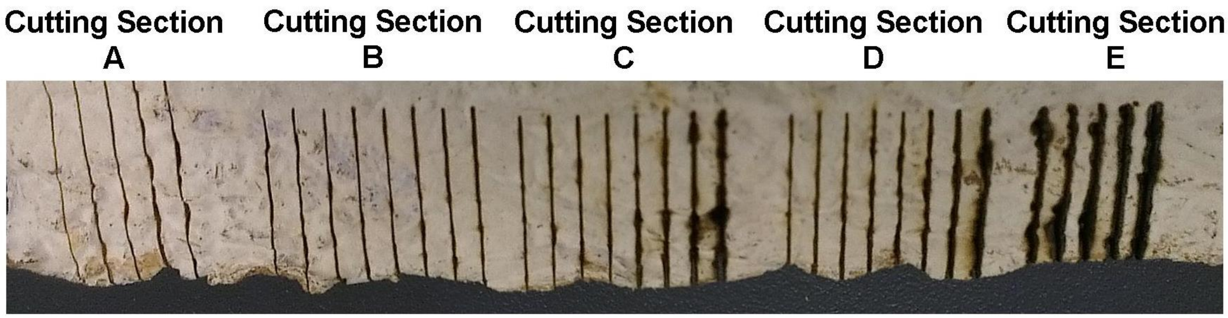

Figure 31. Bulk cutting test results – upper testing surface

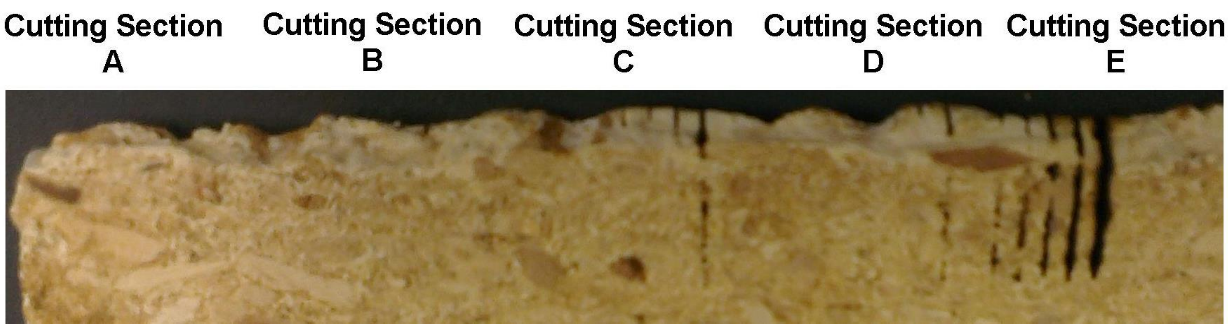

Figure 32. Bulk cutting test results – lower testing surface showing partial penetration

Cutting section A, at 60 Watts with 5% speed, 3.50 inches/second, showed fine upper surface cuts with good dimensional accuracy and precision, but failed to penetrate entirely through the material to the lower surface. There was no visible damage to the material.

Cutting section B, at 60 Watts with 4% speed, 2.80 inches/second, showed fine upper surface cuts with good dimensional accuracy and precision, but also failed to penetrate entirely through the material to the lower surface. There was no visible damage to the material.

Cutting section C, at 60 Watts with 3% speed, 2.10 inches/second, showed upper surface cuts with correct dimensional precision, but with some charring on the last three cuts of the section, likely caused by the heat build-up in the localized area during cutting. This may have contributed to the last cut of section almost penetrating through the material as shown in Figure 32 – Lower testing surface, cutting section C. There was incorrect dimensional accuracy, caused by charring damage, with visible damage on 2 cuts.

Cutting section D, at 60 Watts and 2% speed, 1.40 inches/second (35.6 mm/s) with 1000 pulses/inch (25.4mm), showed similar results to Section C, with correct dimensional precision and increasing charring on the last two cuts of the section, again likely caused by the heat build-up in the localized area during cutting. There was visible damage to 1 cut, and significant damage to 1 cut.

Cutting section E, at 60 Watts at 1% speed, 70 inches/second (1.78 m/s) with 1000 pulses/inch (25.4mm)., which showed significant charring damage to the upper surface on all cuts, with good precision of the cut lines but poor accuracy due to charring. The final cut achieved entire penetration through the material, also showing significant charring on the lower surface.

The bulk cutting results showed that between 3% and 5% speed, 2.10 inches/second (53.3 mm/second) and 3.50 inches/second (88.9 mm/second) respectively, results in partial to full penetration of the material, with visible through to significant damage of the material. The slower the cutting speed, the more energy was imparted to the material by the laser, and the greater degree of charring. Charring was noted in the cuts with full penetration, but may not present an issue for bulk cuts to bring the material quickly to near-net-shape without mechanical damage. Further finishing operations, for example surface coatings such as sealants, would also reduce or eliminate the impact of charring on the final quality and appearance of the material. As laser cutting results in dark colored charring that contrasts to the pale color of the mycelium material, it also makes the process ideal for making purposeful charring and deliberate aesthetic choice through pyrography.

Laser cutting was determined to be a suitable method of subtractive non-mechanical machining, for partial cuts for over 0.12 inches (5.0 mm) thickness, or full cuts for under 0.12 inches (5.0 mm) thickness, for applications where the resulting dark surface finish is acceptable. For under 0.12 inches (5.0 mm) thickness, 60 Watts at 1000 pulses per inch, at a speed of 2.10 inches/second (53.3 mm/second) resulted in acceptable cutting and surface finish performance.

Small Feature Cutting

Precision cutting of mycelium materials is currently limited to mechanical machining methods, which means small features less than 0.50 inches (12.7 mm) maximum size, are difficult to successfully form. This hinders detailed formation of small features on products, and a non-mechanical method would open new options to designers and manufacturers using mycelium materials. Small features have narrow radii and apexes that are not easily formed with other methods, which cause chipping and poor surface finish due to frangible regions in the material. These could be better formed using laser cutting, which can cut to a minimum radius of 4.17×10^{-4} inches (10.6µm), the width of the laser beam. Laser cutting cleaves the material using thermal methods rather than mechanical, eliminating the impact the cutting process has on the surrounding chitin bonds, preventing any chipping damage.

The laser cutting settings for cutting small features was informed by the bulk cutting results, determining the cutting speed required to penetrate and remove the entire depth of the 0.12 inches (5.0 mm) thick sample, to be 60 Watts at 1000 pulses per inch, at a linear speed of 2.10 inches/second for the 0.16 inches (4.0 mm) thick sample. To determine the suitability of laser cutting for small features, a set of shapes were designed. These assessed the formation and performance of forming interior and exterior vertices of a pentangle and square shapes, and of forming internal and external radii of circles, as shown below in Figure 33. The shapes ranged from 0.13 inches (3.2 mm) to 0.49 inches (12.5 mm) in height.

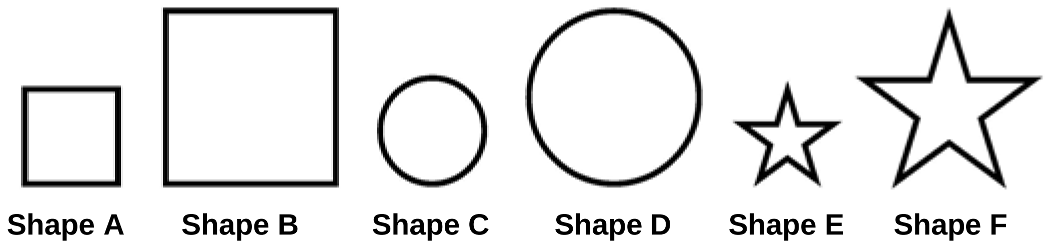

Figure 33. Small feature cutting test shapes A through F

Following the test cuts, inspection showed all of the shapes were cleanly severed from the stock material, and also showed charring but no damage to the cut faces. The small feature cutting test of the vector shapes, shown in Figure 34, showed laser cutting produced well defined edges along the cut surface on both the inner and outer faces of the cut material. The charring damage to the cut faces also resulted in poor accuracy, with the linear straightness of the cut line being affected by the burning of the material. Despite the charring damage, the laser cutting did form sharp vertices, such as for the small and large pentangle shapes apexes. The small scale of the cuts, in the region of ~0.12 inches (~3.0 mm), means mechanical cutting would not be able to form such features, while laser cutting could. The frangible nature of the mycelium material means laser cutting breaks the chitinous bonds without causing chipping, as the charring damage also consolidates the material, by forming a thin surface layer of charring.

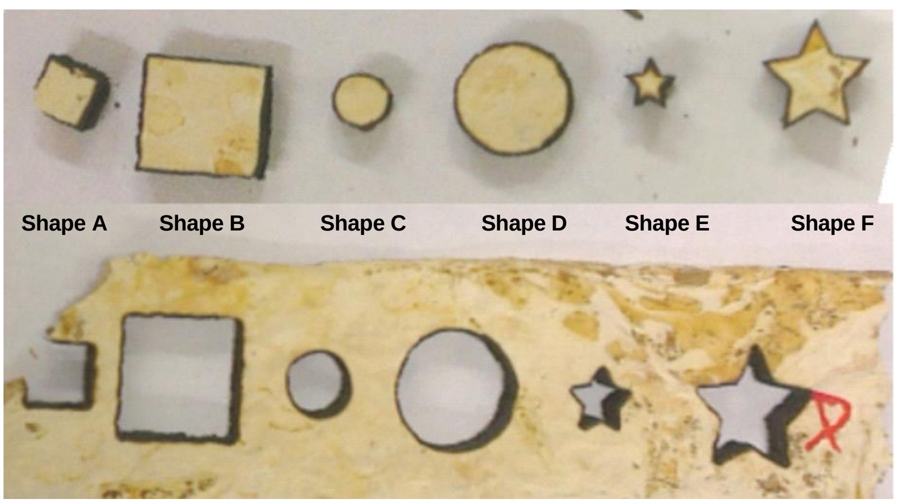

Figure 34. Small feature laser cutting test results
for shapes A through F, showing cutout 'blank' materials (top) and stock materials (bottom)

Shape A, which was only partially cut due to misalignment of the stock material during the test, showed significant charring of the cut faces, and some rounding of the square shape vertices, but still produced recognizable and reasonably well-defined features for a small 0.12 inches (5.0 mm) thickness shapes that could not be cut using mechanical methods.

Shape B showed well-defined vertices of the blank (cut out) piece, but the cutting kerf resulted in poorly defined 90° angles of the square shape on the stock (larger) material.

Shape C and Shape D showed some inaccurate cutting around the circular outline, along with charring damage, but still formed the required shapes adequately.

Shape E and Shape F, with the pentangle apexes forming the smallest features tested by laser cutting of the mycelium materials, showed excellent dimensional accuracy of the blank (cut out) pieces. Some charring damage is seen on the upper surfaces of Shape E, as the dimensions of the piece, 0.13 inches (3.2 mm), approached the cutting kerf of the laser cutting machine, resulting in a charred boarder around the shape.

None of the cutting test resulted in material wastage, producing no chipping of the material or excess material removed from the cutting piece, as laser cutting vaporized any removed material. The laser cutting process also consolidated the cut surfaces through charring, and overall was found a be a suitable method to form small features in the region of ~0.12 inches (~3.0 mm).

<u>Pyrography</u>
Laser cutting methods could provide an alternative to printing or painting finishing processes, as any vector image can be programmed into the laser cutting software. An example would be an organizational logo, providing a fast and single-operation manufacturing solution for forming and finishing at the same time. The surface finish of mycelium materials, being a soft, napped surface due to the biological origins of the material, is not suited to traditional aesthetic finishing operations technologies such as ink-transfer printing or bas-relief machining, as the surface is both textured and frangible. Laser cutting can be used for pyrography, using lasers to burn an image into the material to form a logo or other desired artwork by charring the material surface. This method is suited to mycelium materials as they easily char to form a clear contrast between the black char and naturally light-colored natural surface. The integration of shape cutting and aesthetic finishing could also be an advantage for manufacturers as only one processing step and machine is required, saving time and costs. A 1.5 inch (36.1 mm) Cal logo vector image, shown in Figure 35, was programmed into the laser cutting software, and set to penetrate but not cut the material. The image was chosen to evaluate the performance of laser cutting as an aesthetic finishing operation, due to having complex curves. It was also selected as being representative of a design feature a manufacturer would potentially require as a finishing operation, such as to apply a logo to a product made of mycelium materials.

Figure 35. Laser pyrography test vector image

The samples for the dimensional and aesthetic feature cutting testing were a flat slab of 0.12 inches (5.0 mm) thick mycelium material, selected to provide a suitable volume of material for cutting via a CO_2 laser. The shapes will be visually assessed for precision and accuracy of cut, surface damage and resulting impact on the cut quality and material.

Although the laser beam width is $4.17x10^{-4}$ inches (10.6μm), material is removed by the laser beam during cutting resulted in a cutting beam kerf. To determine the kerf of laser cutting in mycelium materials, the dimensions of the cutout and original material stock after the cutting operation were recorded at several locations, averaged, and compared below in Table 9. The average difference between the cutout and stock material dimensions were calculated to determine the kerf laser cutting can produce on mycelium materials.

The results, as seen in Figure 36 below, showed vector images can be pyrographed into mycelium materials, forming a well-defined image with a high contrast between the material and the intentionally burned areas. The pyrography did not result in excessive damage to the material, beyond the expected burning associated with pyrography. The maximum linear speed of the laser is 70 inches/second (1.78 m/s), with 1000 pulses/inch, suggesting laser pyrography could provide a solution for high-volume aesthetic finishing processes. Laser pyrography was found to be a suitable aesthetic finishing process for mycelium materials, particularly as printing-based processes are not feasible due to the varying surface topology and smoothness. As the process can achieve bulk cuts, it offers the potential for a combined cutting and finishing process for natural materials, without the need for multiple manufacturing steps.

Figure 36. Laser pyrography test result

To determine the kerf of laser cutting in mycelium materials, the dimensions of the cutout and original material stock after the cutting operation were recorded at several locations and averaged, and compared below in Table 9. The average difference between the cutout and stock material dimensions were calculated below to determine the kerf laser cutting produces on mycelium materials. Based on the results, laser cutting of mycelium materials shows an average kerf of 0.04 inches (1.02 mm), with a standard deviation of 0.54. This is comparable to band saw and table saw cutting kerfs, while providing cuts with small detailed features, variable outline cutting paths, and a process that generates minimal to none cutting waste.

Cutting Shape	Dimension Descriptor	Cutout Dimension Average (inches)	Stock Material Dimension Average (inches)	Kerf (inches)
A	Square width	0.19	0.27	0.08
B	Square width	0.49	0.51	0.02
C	Circle diameter	0.23	0.26	0.03
D	Circle diameter	0.48	0.50	0.02
E	Inner pentangle diameter	0.13	0.17	0.04
F	Inner pentangle diameter	0.25	0.30	0.04

Table 9. Initial and final laser cutting pyrography test shape dimensions

Non-Traditional Subtractive Manufacturing Conclusions

Testing showed that laser cutting was determined to be suitable for cutting small features with a minimum feature dimension of 0.13 inches (3.2 mm), and for laser pyrography of vector images. Various examples of laser cut materials are shown in Figure 37. Laser cutting was also found to be suitable to provide a cutting process without the chipping damage caused by mechanical cutting, although the natural constitution of mycelium material results in charring on the cut surfaces.

Figure 37. Examples of laser cut mycelium materials created during laser cutting testing

While full penetration and separation of the material resulted in surface damage through charring, shallower surface cuts proved successful without charring, and could be used for partial cuts or scoring the material. This could be utilized for laminate slabs or sheets of the mycelium materials, to create a flexible sheet similar to the wooden sheet shown in Figure 38, or to create a highly drape-able fabric. Mycelium fabrics, discussed in Section Three, would be an ideal application for thermal cutting due to their structure of thin, laminar sheets. Laser cutting is extensively used for mass-manufacturing within the textile-based fabrics industry (Gao, 2006), and thus would be a suitable choice for larger-scale processes needed for mass-manufacturing of mycelium materials.

Figure 38. Plywood sheet laser cut to form a flexible geometry
(koFAKTORlab, 2013)

Laser cutting small features and bulk cuts also highlighted that the laser cutting process doesn't generate any waste, as the removed material is converted into CO_2 and other combustion byproducts. This could further support the circularity aspect of the material, making a zero-waste manufacturing process to fit with the waste-digesting abilities during fabrication of the material. One consideration of the suitability of laser cutting for mycelium materials is the naturally high surface roughness and varied topography of the materials, resulting in thickness variation which affects the laser penetration. This may have contributed to the dimensional inaccuracy of some of the test cuts. An associated limitation was that laser cutters cannot create five-axis angled cuts, due to the constraint of the laser beam cutting at a fixed 90° the surface of the material, limiting fabrication options. Charring of the materials is also an unavoidable result of laser cutting, which could impact the aesthetics and possibly the integrity of the material at the charred zones. A potential solution could be cutting in an inert gaseous atmosphere such as argon or carbon dioxide, which would prevent or reduce the combustion of the material during cutting.

Given further resources and time, additional testing could be carried out to find the maximum thickness of mycelium material which can be cut using laser cutting. Ultrasonic testing also had good potential to cut bulk volumes of materials, without the mechanical actions that cause chipping and fuzzing. Given the resources were available to continue evaluating cutting processes, testing to identify the resulting surface finish and maximum block sizes that ultrasonic cutting could achieve would be a recommended part of further work. Testing cutting in inert gaseous environments would also provide more information on potential ways to reduce charring and improve the cur surface finish. Finally, while the majority of material properties testing has been carried out with testing samples that have been cut using mechanical cutting methods, exploring the effect of laser cutting on the tensile, compressive and other material properties would also be a recommendation for further work. Mechanical cutting introduces defects such as chipping, which may impact the properties, such as chips reducing the ultimate tensile strength of the material. Laser cutting has been found to reduce and potentially eliminate chipping, which in turn could reduce the impact of defects on mycelium materials mechanical properties. Overall, the laser cutting testing has shown that mycelium materials are suitable to be cut using laser cutting, provided the associated cut surface charring does not interfere with the aesthetics or performance of the final application. Laser cutting was also shown to be suitable to create small, detailed features with a minimum size of 0.13 inches (3.2 mm), and to create aesthetic surface finishes through pyrography.

Chapter Eight – Environmental Degradation Testing

A common question raised by designers when considering using mycelium materials is the performance and degradation of the materials over time. Many applications, such as for architectural and decorative uses, require materials that will not degrade during their use phase. Generally such applications are in interior environments, but mycelium materials may be used in exterior environments, for example in automotive or insulation applications. The mycelium materials could also be used in building construction and residential environments, and may be exposed to exterior or interior conditions. While interior conditions with constant temperature and dry conditions means the material would remain unchanged, the effect of exterior conditions on mycelium materials is largely unknown. The visible surface effects from weathering and degradation are just as significant as the internal changes, as consumer opinions are affected by the visual appearance of a material which infers its quality and performance – an important consideration for a material potentially used in a home or office environment.

To provide designers and developers with information on the integrity of mycelium materials over time, testing was carried out to determine the degradation of mycelium materials in exterior environments. Different types of mycelium materials may perform differently, and so a range of mycelium materials were tested. The molded blocks test specimens of each type of mycelium material were placed in an outdoor environment, attached to a board insulation base, inclined at 45º from the horizontal, facing upward and due south, as specified by ASTM C488−05 Standard Test Method for Conducting Exterior Exposure Tests of Finishes for Thermal Insulation (ASTM C488, 2010). During environmental exposure the test specimens were subject to cyclic temperature change, rain, wind, and natural flora and fauna found in a residential setting.

Fourteen different types of mycelium materials, specified in Table 10, were tested. Specimens were uncoated and left in their natural finish, as this was likely to lead to the lowest performance and thus the most accurate determination of the materials lower-bounds of performance under normal use conditions, which coatings, sealants etc. could improve on. For comparative purposes, a set of materials from the same batch of each type were placed in an identical arrangement in an indoor environment, to act as controls.

Prior to exposure, material types B1, B2, C, F, G, H, I, L and M all showed a white/light yellow chalky soft napped texture. Types A and K show an unfinished soft wood like surface. Types D and J show plywood and bamboo laminate surfaces. Specimen D was laminated with plywood layers of 5.00 mm (0.20 inches). Specimen type J was laminated on the upper and lower surfaces with bamboo layers of 3.50 mm (0.14 inches) with wood glue. Both specimens were left uncoated in their natural finish, a light beech colored smooth wooden surface with matt texture. Type E showed a semi glossy hard wood like surface, being a denser type of mycelium material. The specimens showed no evidence of discoloration, cracking, crazing, flaking, shrinkage, blistering, holes, or delamination. There was light to moderate chalking on types B1, B2, C, F, G, H, I, L and M, which is one of the natural surface finishes of de-molded and desiccated mycelium materials.

Type A – De-skinned *Ganoderma lucidum*	Type G *Ganoderma lucidum* with hemp feedstock
Type B1	Type H

Assorted species Higher density	*Ganoderma lucidum* Higher density
Type B2 Assorted species Lower density	Type I Assorted species
Type C *Laetiporus sulphureus*	Type J Bamboo laminated *Ganoderma lucidum* core
Type D Plywood laminated Ganoderma lucidum core	Type K *Ganoderma lucidum* Medium density
Type E *Ganoderma lucidum* extra dense	Type L *Ganoderma lucidum* Lower density
Type F *Ganoderma lucidum* with thickening additive	Type M *Ganoderma lucidum* with husk feedstock

Table 10. Mycelium material types for exterior exposure tests

ASTM C488−05 Standard Test Method for Conducting Exterior Exposure Tests of Finishes for Thermal Insulation (ASTM C488, 2010) was used to determine how the mycelium materials change and degrade in exterior conditions. The test site selected was a residential area near Berkeley, CA, USA at 37.8700° N, 122.2590° W, at an elevation of 52 m, chosen to represent a typical mild temperate environment in a residential building setting in which the mycelium insulation material could likely be used. The average climate of the region during the testing period was a maximum temperature of 70ºF (21ºC), a minimum temperature of 49ºF (9ºC), and 0.87 inches (22.0 mm) of precipitation. The test specimens of each type of mycelium material were molded in blocks of 3.37 inches (85.5 mm) length, 2.23 inches (56.6 mm) depth and 0.93 inches (23.5 mm) thick, with the exterior skin of thickened mycelium created by contact with the impermeable mold surface, left intact. The test specimens were attached to a 2.95 inches (75.0 mm) by 1.97 inches (50.0 mm) board insulation base, inclined at 45º from the horizontal, facing upward and due south, as specified by ASTM C488-05. The test specimens were arranged on the insulation base, and exposed to the exterior environment for 7 days, with visual inspection and image recording on the first and last day. They were examined for signs of evidence of discoloration, cracking, crazing, flaking, chalking, shrinkage, blistering, holes, or delamination, in comparison to unexposed control set of materials. The control materials which experienced an indoor environment showed no changes.

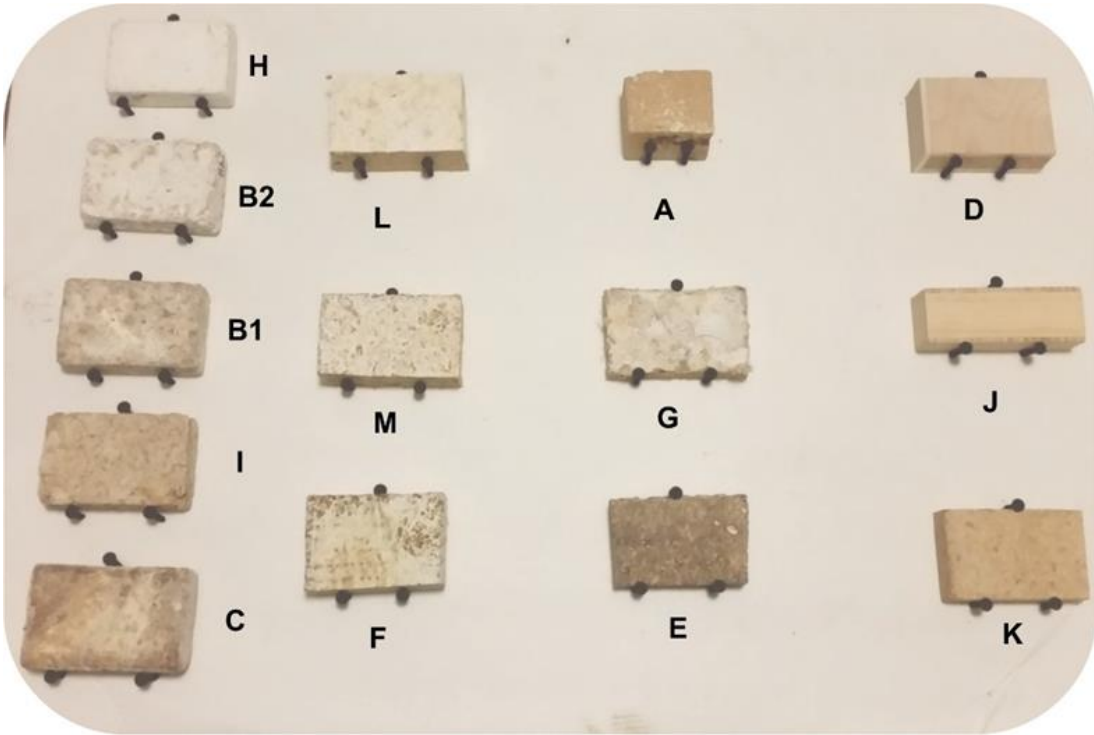

Figure 39. Test specimens before outdoor exposure.

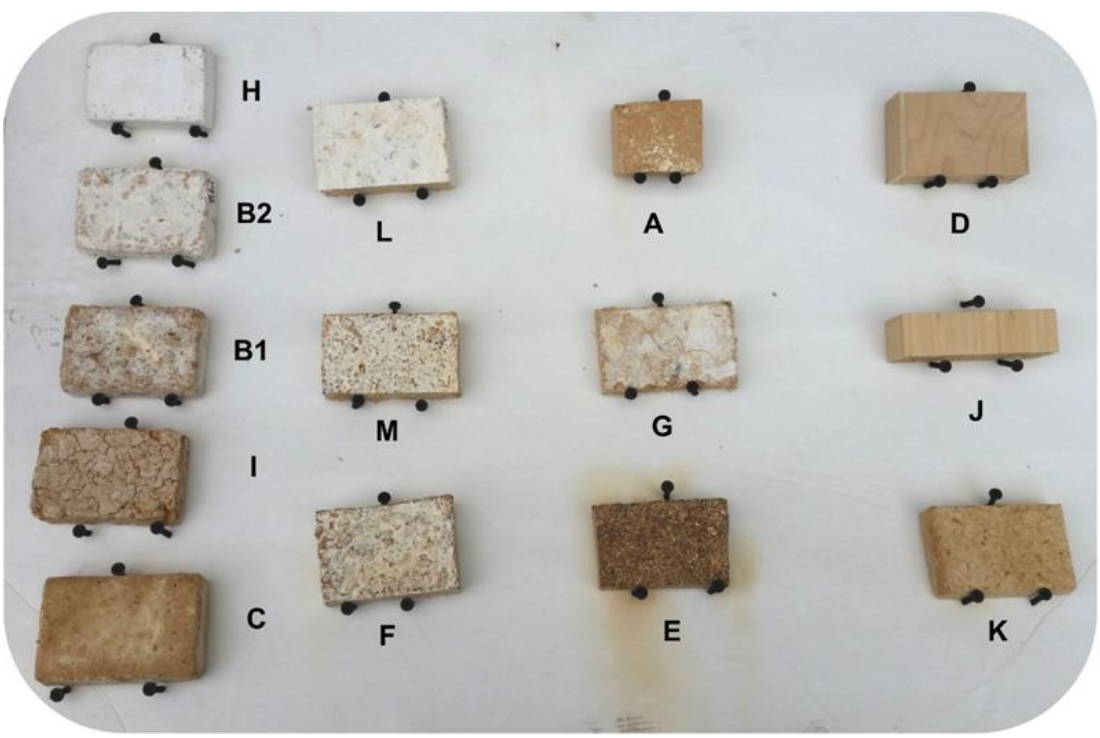

Figure 40. Test specimens after outdoor exposure.

After exposure, as seen in Figure 40, overall the materials showed little changes or degradation. After exposure material types A through C, and F through M, showed no signs of discoloration, cracking, crazing, flaking, shrinkage, blistering, holes, or delamination.

Prior to exposure there was light to moderate chalking on types B1, B2, C, F, G, H, I, L and M, which is a natural finish of the mycelium materials exterior skin. After exposure, less chalking was seen, likely due to the 0.25mm (0.01 inch) rainfall during exposure washing the samples surfaces and removing some of the natural chalking material.

Material type E, which comprised of a higher density material from the same fungi species *Ganoderma lucidum,* showed signs of discoloring the facing board as seen in sample E of Figure 40.

As material types D and J were laminated with plywood and bamboo respectively, there could have been greater potential for moisture damage or delamination, a common problem with synthetic composite materials. However, types D and J the plywood and bamboo laminates, did not show significant signs of delamination or degradation of the open surfaces.

In summary, The effect of exterior exposure on the mycelium materials, blocks were placed in an outdoor environment for 7 days, facing southerly and in a mild temperate environment of minimum 9ºC (49ºF) to maximum 21ºC (70ºF) with an average rainfall of 0.01 inches (0.25 mm). Overall the materials showed little to no changes or degradation during exposure – the mycelium materials were quite robust, with weathering similar to unfinished wooden materials. None of the material types showed damage from fauna such as insects, though longer-term testing would be needed to fully determine the mycelium materials' resistance to pests and insect damage. It should be noted as a natural material, mycelium materials are predisposed to degrading in a natural environment. However, this requires biological vectors to be present such as contained in soil, along with moisture, in order for the material to degrade. Relatively there were limited changes to the material during the exposure time, with weathering similar to unfinished wooden materials, and withstood 7 days in an exterior environment. While the mycelium materials were clearly not waterproof and thus best suited to an indoor environment, they were tested in their natural finish and unsealed. Exterior applications would be possible by adding sealant to prevent water absorption would prevent the degradation of the materials, which are desiccated during their fabrication process. If the mycelium materials were sealed, using a natural varnish such as shellac, natural sealant like latex, or a synthetic coating such as epoxy resin, they would be suitable for outdoor environments. The lack of any change to the control materials in an indoor environment also meant they are suited to indoor applications. For example, as a 100% recyclable material made from sustainable waste agricultural feedstocks, mycelium materials are comparable to fiber glass insulation but non-toxic. Having been shown to be suited to indoor environments, they could be an alternative to current home insulation materials.

Chapter Nine – Fungi Materials as a Biological Technology

The species selection, growth conditions and time, and post-growth processes are all key aspects of forming the mycelium materials into useful products. Careful selection of the fungi species allows for various material and growth properties, with this research being focused on mycelial growth of the *Ganoderma lucidum* species. Each of these key factors are briefly explored, to provide insight and background to the fabrication of the materials used in this research.

<u>Species Selection</u>

There are over 5 million known species of fungi (Blackwell, 2011) many of which many are edible, and grow in symbiotic relationships with other biological organisms including trees. They have a wide range of biological, mechanical, and growth properties. Selection requirements for choosing the species to cultivate for research included factors such as:

- Fast growth, compared to other eukaryotic organisms (Cavka & Jo, 2014)
- Safe for humans while in the pre-fruiting stage (including being edible to reduce any risk of accidental ingestion)
- Ability to adapt to feedstock requirements
- Ability to grow in easily reproducible environmental conditions close to standard pressure and temperature conditions, also including humidity, and the pH of water required to grow
- Visible light intensity needed for growth.

The species selected for the growth of the mycelium material has a significant impact on the final material type and characteristics, as each species of fungi has unique growth patterns, vegetative structures and resultant properties. Much of the research into the properties of fungi has been completed within communities of fungi enthusiasts, or from a niche industry such as edible mushroom farming, and data is often hidden and unavailable within the public domain.

Fungi are neither a plant nor an animal but have features of both, including being eukaryote cells which have membrane bound organelles. Despite being genetically more similar to animals than plants, fungi have their own classification kingdom, due to having cell walls which contain chitin which differentiates them from plants and bacteria. Fungi are typically the recyclers of nature, principally decomposing matter using digestive enzymes and are often overlooked due to their cryptic lifestyle, meaning their growth is often hidden from view in soil or dead matter. The more noticeable and typical symbolism of fungi is their fruiting bodies, often grown for human consumption, which also provide part of the reproductive mechanism of fungi. Several phyla, or divisions, of fungi have been classified including *Basidiomycota* which are fungi composed of hyphae, the long branching filamentous structures (Ainsworth et al., 2006). Selection of the species used to grow mycelium materials can be informed by understanding the growth structures that result from the different phyla. The resulting hyphae structure, skin formation, ease of use, and positive health associations were all factors in the selection of the species *Ganoderma lucidum* for the mycelium materials explored in this research. *Ganoderma lucidum* was also selected for testing due to being safe for human consumption, eliminating the risk of potential harm from accidental contact during testing. *Ganoderma lucidum* is also known for its glossy, chitinous, skin-like surface formed of the upper skin of the fungi, which along with its hyphal-based structure, make it an excellent choice for use in growing mycelium materials. *Ganoderma lucidum* is known by many names, including Lingzhi, literally “supernatural mushroom” in Chinese, and Reishi “sage mushroom” in Japanese (Paterson, 2006). These naming connotations come from the known health benefits of ingesting this species, including anti-inflammatory activity by inhibiting lipopolysaccharide induced nitric oxide production in macrophage-like cells (Geng, 2014) (Hasnat, 2014), anti-cancer properties (Martínez-Montemayor et al., 2011) (Wang et al., 1997)

(Min et al., 2000) potentially due to complementary adjunct treatment by improving immune function (Jin et al., 2012), along with several other health benefits for diabetes (Ma, 2015) and gastrointestinal health (Chang, 2015). *Ganoderma lucidum* has also been linked to anti-HIV therapies (El-Mekkawy et al., 1998), and although the potential health benefits of the fungi used in this research is outside of the scope of this research, it is noted to give some background on the potential for these materials to have more than one function. This increases the likelihood of consumer acceptance of the material as the species already has a commercial presence being bought for health purposes, a useful characteristic for consumers' commercial perceptions when used as a material – very few furniture or low-load construction materials are edible & have health benefits.

<u>Biological Materials Growth</u>

The initial stages of growth of mycelium materials were completed in growth bags with a specifically designed patch to allow the exchange of oxygen and carbon dioxide for the fungi's respiration and growth, without allowing the egress of biological spores, or ingress of competing fungi species. One of the major challenges in the growing of mycelium materials is the need for near clean-room conditions, as the feedstocks and growing environment provide perfect growing conditions for a variety of fungi species and airborne pathogens, all of which can geminate and become competing species with the chosen fungi species. Following inoculation of *Ganoderma lucidum* into a mix of *Quercus rubra* (commonly known as red oak) feedstock, nutritional supplements, and a pH buffer solution, growth was continued for approximately 14 days at 23ºC (73.4ºF) in nominal indoor daylight conditions. Each fungi species has a required temperature of ideal growth, and unlike synthetic materials such as polymers, these are typically around standard pressures and temperatures. This is an advantage if growing fungi is scaled up to mass manufacture volumes, as production processes that do not require large amounts of energy (and associated costs) during production are inherently more sustainable in a wider context, than materials which require high energy use during manufacture.

Fungi growth is governed mainly by nutrients, temperature, light, aeration, pH and water activity (Litchfield et al., 1963). Water is the most important feedstock required, with the species *Ganoderma lucidum* requiring excess water, in which the acidity levels and activity (physical excitation) of the water both play a role. While the latter factors are well known, even if specific values of volumes and pH are not, much of this information is proprietary and is not available for publication within this research. Aeration is key for gaseous exchange of oxygen and carbon dioxide – aeration has proved the most common source of contamination with competing species, despite the need for free exchange needed for growth. Light can be a negative growth factor, as it triggers the mycelium to increase growth of chitin, creating a hard shell of impermeable biological material to protect against moisture loss and exposure to too much light (Stajic et al., 2002). A factor rarely described in research into fungi growth, excepting fungi as a farmed or foraged food source, is the length of growth time and resulting associated properties. Similar to a synthetic composites, mycelium materials contain both a matrix, made of fungi, and fibers from the original cellulosic feedstock. The composite begins growth entirely composed of the fiber of the feedstock mixed with spores of the fungi, and during the digestion and growth process the feedstock is converted to the mycelium matrix, achieving an end mix of feedstock fibers and mycelium, in this case a composite of *Quercus rubra* and *Ganoderma lucidum* respectively. The length of time required to digest the feedstock, and the resulting density, strength etc. of the material has largely been determined experientially and somewhat in a trial-and-error fashion, again with much of the information being proprietary and unavailable. Nonetheless, anecdotal observations during the course of this research has found the finer the feedstock, and higher the resulting density of the mycelium material. This is theorized as being due to forming a higher density of hyphae around the high density of available feedstock, resulting in a dense mass of hyphae which creates a dense final material. This warrants further investigation to determine if this is common to all mycelium materials, or just ones based on *Ganoderma lucidum*, but due to the time and resource constraints this was beyond the purview of this research.

The type of feedstock nutrient, dispersion of and feedstock particle size are all factors in the breakdown of lignin containing materials (Buswell et al., 1993) into the resulting mycelium composite's composition and resulting properties. During growth time, the fungi species increases in hyphal mass, seeking nutrients, water, and exploring its surrounding using the hyphae. The current theory developed from observations on the geometry of samples during growth phases, is that the fixed volume of the mold encourages increased density of the mycelium hyphae, whereas growth unlimited by volume results in less homogenous and less dense mycelium growth. This trend and the factors affecting such growth is still largely unknown would be an excellent avenue of further research.

Once the required degree of growth is reached, the materials are removed from the molds, having formed blocks of mycelium material mixed with the remaining feedstock fibers. The de-molded blocks, shown in Figure 41, show an approximate 0.0394 inch (1.00 mm) thick encompassing layer of pure mycelium material, seen visually as a white layer surrounding the block. This layer develops in reaction to contact with the impenetrable mold interior.

Figure 41. De-molded block of mycelium composite, showing white mycelium surface growth

Growth Factors & Impact on Fungi Growth

Directing the growth of mycelium materials can be achieved in a variety of ways, most having been identified in a practical, and sometimes trial and error, context during commercial development efforts. Each of the following factors that go into growing mycelium materials affect the resulting form, consistency and physical properties of the resulting material.

- Species type
- Feedstock type
- Feedstock size
- Availability of feedstocks
- Sterility & competing organisms
- Growth time
- Mold material
- Temperature
- Humidity
- Aqueous level
- Light level

Directing growth can be an active or passive method, active being a constantly applied situation that over-rides or prevents growth (such as a physical mold), and passive being providing favorable conditions that the fungi choose to follow (such as controlling feedstock location and availability). Unlike plants fungi do not need light to grow (Tisch & Schmoll, 2010). Light can be a negative growth factor, and Bayer & McIntyre (2011) also noted light direction, intensity and diffusion can be used to control growth direction. Fungi choose their direction of growth using a variety of methods, and during the fruiting stage use growth direction to orient themselves and their fruiting bodies. This could be an avenue to control the formation of the material, without needing additional inputs. An example could be rotating a mold to result in equal growth in all whole volume. Fungi have a negative tropic response to gravity, gravitropism, which is used by fungi to determine the direction to grow that is away from Earth's gravity. When oriented for fruiting by releasing spores their gravity-based spore dispersing membranes called hymenophores, to prevent spores being trapped as they exit the membranes and reducing spore dispersal. Fungi's gravitropism was confirmed when fungi were grown in zero gravity. With no gravity force to signal the growth direction, the fungi grew directly away from their growing media and irrespective of the current 'vertical' direction. Cultures of *Flarnmulina* were flown on the Spacelab D-2 mission in 1993, the resulting disorientation of fungi fruit bodies proved fungi's 'gravitropism' response is due to the unidirectional gravity vector of Earth (Moore et al., 1996). The mechanisms which govern growth are complex, and for this work are only explored in terms of ideating a potential way to control fungi as a technology, and so further detail on the growth mechanisms are not covered in this body of research. Commercial mycelium materials are stopped growing before they reach the fruiting stage, so this method of growth control would only be practical if the fruiting stage was controlled and delayed. Since the factors described above have to be considered when growing and fabricating mycelium materials, it is unknown how they are applied in many commercial applications due to being trade secrets. Despite this, some information is available from academic and marketing sources such as websites from commercial ventures. While entrepreneurial activities do not routinely publish information, except through academic collaborations (such as this research effort), the majority of information that is shared has the goal of informing and supporting adoption of mycelium-based materials. The following sections use this information to summarize the main fabrication and denaturing processes mycelium materials developed so far.

Mycelium Materials Fabrication Process

Mycelium materials can be used to form the matrix of biocomposites, and can also be reinforced with natural fibers. Due to the fact that the morphology of fungi changes during the different growth stages, and chitin can form into allotropes with different properties (Tang et al., 2015), mycelium composites can be developed to form laminate morphologies during their natural growth stages. Fungi grow by forming complex cellular structures, with varying formations of expanding cells called hyphae, to form a network of mycelium which naturally occur as the fungi grows and seeks food. With qualities of both plants and animals, as fungi grows it expands and uses its hyphal networks seek out nutrients and transport them within its body. Hyphae typically form branching structures as they grow, and unless given unlimited nutrients, will form a consistent density. Varying the nutrients and growing media can produce specific growing morphologies (Edelstein & Segel, 1983), which can be utilized for various specific applications. Since there are a huge number of fungi species, and more are still being discovered. Each of their responses to their growing environment and stimulus is different, fungi represent a high potential for finding an entirely natural material which can fulfill a multitude of roles for material applications.

During the growth phase the physical shape and geometry of the mycelium materials can be configured to produce the final near-net shape desired. This enables the use of molds of form the desired near-net shape, with the feedstock filling the mold cavity and the fungi digesting it to form the biocomposite in the form of the mold. Different geometries and consistencies can be created using various molds and molding processes, including variable exterior skin morphologies and thicknesses. Due to the natural growth of the fungi species, molds must have open access for gaseous exchange during the growth phase, and also be made of an inert material resistant to digestion by the fungi species. If no specific geometry is required of the material, the mold can be flexible sided polymer bags, as shown in Figure 42.

Figure 42. Mycelium materials in growth phase

Growth of mycelium materials in molds results in variation of the skin on the mycelium in contact with the mold. As a naturally growing organism, the *Ganoderma lucidum* culture reacts to its surroundings, including forming natural barrier skins to impermeable surfaces, such as the mold container. Inspection showed the chitin layer naturally thickened when in contact with the impermeable and impenetrable mold material, to protect the growing fungi and to prevent moisture loss. The growing conditions for the *Ganoderma lucidum* based mycelium materials are best achieved in a controlled environment at room temperature, around 20ºC (68 ºF), with wood or other specified feedstock and sufficient nutrients for a period of approximately 14 days. Compared to similar natural materials such as cork and wood, which take many months (and in case of hardwoods, years) to grow, mycelium materials have a fast growth rate. This is a key advantage of mycelium materials over both synthetic and other natural materials, as a fast growth rate is important if they are to scale up production of mycelium materials to the volumes needed for mass manufacturing. This means manufactures could produce large volumes of materials quickly. Additionally, unlike synthetic and other (and sometimes limited as in the case of natural cork) natural materials, scaling to greater production volumes of mycelium materials does not result in equally greater use of energy resources. This is because they are grown from waste and typically grow at room temperatures and pressures, also without the need for light sources, reducing the costs and resource demands of scaling up production.

The basic mold shape for the mycelium material samples was approximately 85 mm by 55 mm by 30 mm, although a variety of mold shapes are possible. The fungi species digests carbon-containing organic materials, such as wood chips, to grow a network of hyphae. Hyphae are individual strands that form the vegetative growth of the fungi, and together form the mycelium that make structure of the fungi (Stamets, 2005). Figure 43 summarizes the mycelium materials' fabrication process.

Figure 43. Mycelium materials fabrication process overview

Due to the growing conditions also being favorable to many other natural species of fungi and various competing organisms, the growing conditions must be treated carefully to ensure the culture is purified so only the selected species grows. Control of the growing conditions is equally important to develop the required consistency of the material, due to the fact that the cellular material has different densities and structures in response to different conditions, growth times, and feedstocks. As the fungi digest the carbon and other organic molecules and convert them into the chitin cellular structure of fungi, the choice of feedstock can be used to control the resulting properties of the material. The current mycelium materials used for bulk applications, such as packaging, use wood-based feedstocks rich in lignin. Since each species of fungi is specialized into growing optimally on a particular feedstock, a variety of feedstocks can be used depending on the species selected.

Bayer & McIntyre (Bayer & McIntyre, 2011) noted that fungi also self-digest. They can also be joined during the growth stage. The two blocks of the mycelium materials are sanded to a depth of four thousandths of an inch (0.1 mm) and placed in close physical contact, and enzymatic signaling occurs which triggers growth, combining them into one single continuous block. This could provide a way to combine and adhere blocks together without the need for adhesives, suggesting a way to combine large quantities of the material without the cost or resource demands for glues.

<u>Denaturing and Post-Processing</u>

At the microscopic level, fungi grow in a somewhat aqueous environment, depending on the species, with the hyphae extending through the wet growing space and voids. As a live organism, once the growth phase of the fungi is complete and the required growth and characteristics have been achieved, the fungi organism must be denatured to prevent further growth and development. This is also required to ensure the fungi species does not continue growth into the reproductive stage, which would produce reproductive spores which can cause allergies in humans (Gravesen, 1979).

Denaturing organisms requires heat processing to above 158˚F (70˚C), which disables cells functions by denaturing the cell proteins (Rosenberg et al., 1971), rendering the mycelium material lifeless. Since the material is denatured prior to the fruiting which fungi species use for reproduction, thus the final material contains no fungi spores and is entirely inert. The heat process, typically achieved in an industrial oven, also serves to desiccate the mycelium materials to around 5% moisture content as measured with a digital moisture meter. This also helps prevent future growth of the fungi, and of other flora or fauna which could also feed on the natural mycelium material. There are a variety of post-growth processing options for mycelium biocomposites, including reprocessing and re-growth, and the application of natural exterior sealants, such as shellac or natural latex.

Creating barriers to mycelium during growth of the fungi is also a possibility in forming the material into useful shapes. Recently, a project by Dr. Rooth and Dr. Johansson at Nanexa, a company focused on nano-based drug delivery, found mycelium could be prevented from growth into polymeric materials such as polyethylene and silicon. This was achieved by creating a layer of silver particles, which naturally denatures fungi (Melaiye & Youngs, 2005). This means that molds with a silver & silicon coating could be made from polyethylene materials, rather than the current metal or thermoset materials currently used, suggesting recyclable molds could be created to further lower the impact of the materials.

Materials Safety & Ethics

The common question when presenting this research, is "is this safe?". As mycelium materials are grown from the living tissue of fungi, which is denatured before it reaches the fruiting stage, they are inert. This is to prevent the fruiting stage of fungi, which produces fungal spores, the reproduction parts of the fungi, which are hazardous to human health.

Additionally, using species of fungi that are edible (and therefore safe even during the growth process), creates another dimension of safety for the material, particularly if entirely natural feedstocks are used. Finally, so far fungi have not been found to have self-awareness, but do make complex decisions about their growth and survival strategies – future research should be mindful of safeguarding the ethical treatment of fungi, if they are shown to require the same ethical safeguards as animals.

Section Two Summary: Fabrication, Forming & Safety Similar to Wood Materials

To understand mycelium materials behavior and suitability for useful applications, the fabrication and manufacturing processes required to form the materials from waste feedstocks were considered. The empirical research found basic features can be formed using mechanical cutting methods, such as drilling, milling and sanding. However, due to the frangible nature of the material, detailed features with a minimum size of 0.13 inches (3.2 mm) were best formed by laser cutting. This also produced charring of the material, which could be exploited as pyrography, to create surface decorations such as logos. Together, these processes provide a range of methods to cut, form and detail mycelium materials, best accomplished using tools appropriate for wood-based materials and laser cutting. A range of mycelium materials were found to suffer minimal degradation during outdoor exposure conditions, and no degradation during interior conditions. The key fabrication, denaturing and post-processing of mycelium materials were also identified. Based on the results of Section One and Two showing the properties of mycelium materials make them suitable for a range of applications, current and developing applications for mycelium materials are explored in Section Three.

SECTION THREE: APPLICATIONS

Fungi serve a vital role in all ecosystems and have native species present in every environment and soil type on the planet, even deserts (Ranzoni, 1968). Along with being nature's recycling mechanism, the deeply complex and varied relationship of fungi with various plants, animals and even in the human digestive system (Hallen-Adams & Suhr, 2017) means they have been developed for useful applications in a wide variety of ways. As there are millions of fungi species (Hawksworth, 1991), each with its ideal environment, feedstock and resulting useful properties or applications, including just a few use cases listed below:

- Food and beverages – Fungi, particularly the *Candida* and *Brettanomyces* genera are used as yeasts for brewing and bread making (Skinner, 1947).
- Fertilizers – Fungi form symbiotic (symbiont and host) & mutualism (optional, mutually beneficial) relationships with plants – examples include mycorrhizal root relationships (Gianinazzi-Pearson, 1996).
- Natural waste remediation – Coprophilous (literally "dung-loving") fungi (Thilagam et al., 2015).
- Soil remediation – Removal of heavy metals and bacteria from soil using fungi (Khan et al., 2000).
- Construction materials – Bricks (Critical Concrete, 2018), insulation, non-structural interior wall panels, and decorative materials (Holt et al., 2012b)
- Vibration damping and acoustic absorbing materials (Holt et al., 2012c)
- Clothing, including hats, shoes (Jiang et al., 2014), (Silverman, 2018) and handbags (MycoWorks, 2016)
- One-use-cushions for tsunami buoy deployment (Holt et al., 2014a)
- Medical uses including skin grafts and even anti-cancer properties as a dietary supplement (Sliva, 2003), (Su et al., 1997)

Due to vast scope of the fungi-based technologies, only recent developments related to fungi as a material were considered in this research, and mature technologies such as brewing, medical, and purely biological aspects of fungi are not considered in this research. As some of the applications for fungi materials are long-range options, only likely to reach commercial maturity in the future, the case studies do consider fungi materials from a slightly wider scope to include sanitation/remediation applications.

Sharing learning and challenges between fields would help develop the use of natural materials, including mycelium materials. The wider industry of natural materials is still an emerging trend, constrained by the higher costs and more specific processing requirements, but continued pressure from consumers to understand and control the impact of their products slowly drives growth in the field. Newer technologies are still being developed, and have the potential to reduce waste and our impact. This means researchers should continue to characterize mycelium materials, and find more innovative applications. Circular are an increasing trend within business and industry, seeking ways to reduce impact without additional cost or reduction in quality. An example is Apple's recent moves towards recovering smartphones from consumers, dissembling them, and reusing various materials to produce more Apple products. Significant savings can made from the reduction in costs of purchasing increasing expensive and rare materials from around the world, switching instead to investing in materials as a circular technology. While this is currently based on high value metallic and rare earth elements, biomaterials can offer the same circular concept, but with lower-energy materials that also remediate wastes from other processes or products. Currently chitin-based mycelium materials are a growing option in the natural materials field (Harish & Tharanathan, 2007), (Rinaudo, 2006), and (Kumar, 2000). Several artisanal furniture products have been grown from mycelium materials. The material has also been commercialized into packaging by New York based firm Ecovative Design LLC, and has been developed for use by large-scale buyers, including by the worldwide furniture manufacturer IKEA (Gosden, 2016).

The following Table 11 summarizes some of the current and potential applications of mycelium materials. It indicates the current materials used for various applications, the key properties that these materials are chosen for, relative cost, and the issues or shortcomings these materials suffer. Since these shortcomings are in turn opportunities for mycelium materials to provide solutions, mycelium materials ability to replace such materials is rated from low to high. This is determined by considering the likely ability to them to deliver the required properties, become cost competitive (as far as possible within economies of scale), and ability offer consumers advantages the current material does not.

Application	**Material Requirements**	**Typical Materials**	**Relative Cost**	**Material limitations**	**Mycelium Materials Potential**
Packaging	Lightweight Recyclable/compostable Low toughness	Expanded poly-styrene	Low	Polluting Contains carcinogens	High
Home insulation	High thermal insulator Lightweight	Glass fiber	Low	Few disposal options	High

		Themoset resins	Low	Contains carcinogens	
Wall panel	Lightweight High compressive strength High stiffness	Wood & expanded polystyrene and polyurethane foams	Medium	Few disposal options	Medium
Furniture e.g., chairs	High compressive strength High durability Good machinability	Hard woods Polypropylene	High Low	Limited resource Non-recyclable	High
Wine stoppers	Elastic Waterproof	Cork	Medium	Limited resource	Low

Table 11. Applications for potential materials replacement by mycelium materials

While many applications currently use synthetic materials, despite offering low materials costs, they are also made from crude-oil sources. This means they typically have very limited recycling options, resulting in a relatively high impact on the environment. For example, chairs are often made of polypropylene, which is a durable, tough and relatively low cost polymer, but has limited recycling or reuse options. Despite this concern, most consumers and manufacturers are more sensitive to the cost of materials than the environmental impact, and choose lower-cost, mass-produced synthetic materials despite their worse environmental impact. Mycelium materials have a good potential as they use waste as feedstock, suggesting the potential to bring down the cost of the material, by offsetting the production costs with a free (or even cost negative) feedstock such as sawdust or waste polymers. This also provides the unique selling point of the mycelium materials as a less environmental harmful choice. If reduced environmental impact is key consumer consideration or unique selling point of the product, then higher costs are less of a barrier to purchase to consumer (D'Souza, 2007). If mycelium materials can become more price competitive, it could become a viable option to replace synthetic materials.

Another key attribute of mycelium materials is their ability to have their properties customized. For example, they can be grown as lightweight, low-strength materials, or high-density, higher-strength materials. Unlike other natural materials, such as softwoods like balsawood, mycelium materials properties are dependent on the feedstock, growth conditions & time, and can also be combined as sandwich composites to meet a wide variety of materials properties. For example, wall panels for use in home decorating require both lightweight, compressive strength, and good surface finish to paint upon. Combining low-density mycelium materials that have a high thermal insulation value, with higher-strength bamboo, results in a sandwich composite able to offer similar properties to the current panels containing polystyrene. Similarly, for applications that require waterproofness (not an intrinsic property of mycelium materials due to being desiccated), the addition of latex, shellac or other natural sealants can make it suitable for a wider range of applications.

To provide examples of mycelium materials used in current applications, and explore future applications, I completed three case studies. Two of the case studies focus on current and developing applications, and a third introduces a potential application for mycelium materials as a sanitation technology to be used in low-resource conditions.

Application Case Study Methods

To study how mycelium materials have been developed and commercialized, and the suggested trajectories of such efforts in the future, two different research methods. These methods were chosen to suit the purpose of understand three very different representative applications out of the wide range available. The three selected applications – packaging, leather replacement material, and sanitation – were selected to give examples of the range of development stages of mycelium applications that exist. Packaging has already developed and commercialized, leather replacement materials are currently in development but not as yet commercially adopted, and sanitation is a novel conceptual application, neither developed nor commercialized as yet.

The first application, packaging, was explored using an illustrative case study, a type of case study which serves to bridge the gap in the understanding of a topic to a wider audience, and provide a common language with which to discuss the topic (Hayes et al., 2015). This method was chosen to provide an overview of mycelium materials in its applications, limited to a scope and depth appropriate to familiarize the reader with the key information and developments in the field. The second application, leather replacement material, focuses on using mycelium materials as a biomimetic replacement for leather materials. It uses a more in-depth and technical method, including analyzing scanning electron microscopy images to give a technical view of the materials, their morphology, and commercial viability as an alternative to leather.

While the first two cases are primarily descriptive studies, the third case study on sanitation has an additional, more in-depth literature based research aspect, to address the novelty considering fungi's aptitude to be a sanitation technology. Initially, the goal was to complete a practical project testing out the concept of fungi used as a sanitation technology. However, due to time and resource constraints, the concepts are outlined instead. More information on the barriers and challenges of this research is discussed in the next section.

Barriers & Challenges

This research has shown several challenges and barriers to development in exploring the range of applications, and determining the role of fungi-based technologies from material replacement and sanitation aspects. Beyond a brief and conceptual level, the complexity of considering mycelium materials as a sanitation technology could have benefited from a longer and more focused research period. While the case study identified the main development points, sources of information and similar commercial entities, ideally a study project to prototype the technology could have been used to determine its feasibility and impact. This would be carried out a low-resource environment, where novel sanitation technologies are needed due to a lack of sanitation infrastructure. While the initial aim of this research was to include the project, after completing the case study the scale and scope of doing such a project within the time and resources available was not feasible, so the case study work was included to be of use to future researchers. Due to the concentration of expertise in development engineering at UC Berkeley, similar research efforts have been made for creating technology to improve sanitation in low-resource areas that also creates useful products. A recent example of this was the start-up Sanivation, which turns urine into fertilizer from compost toilets. Connecting to these resources would help support the project, and understanding key technical, regulatory and cultural requirements of sanitation technologies. Part of designing an effective fungi-based technology for use in low-resource environments, would be focusing on user-focused design, ensuring the toilet is adapted to, and adopted by users. Finding more stakeholders, and continuing work with sanitation experts at UC Berkeley, would help add resources to the project.

Chapter Ten: Current & Developing Applications

The first two case studies focus on use cases of mycelium materials that are currently commercially available, or currently in development within a commercial organization such as a start-up. They demonstrate how mycelium materials can replace other materials, both synthetic and natural, due to offering similar properties and also reduced environmental impact.

Case Study One: Sustainable Packaging

The motivation for studying packaging was due to it being the most commonly produced mycelium-based material currently available. This fits with the mechanical and thermal testing results explored in Section One, which found mycelium materials as being suitable replacements for polymer foams, such as polystyrene packing peanuts. An example is Ecovative Design LLC's packaging materials that are made from agricultural feedstocks and fungi, which are designed to provide short-term impact and thermal protection for products such as when shipping wine bottles, is seen in Figure 44 and Figure 45.

Figure 44. Ecovative Design LLC's fungi-based materials designed for wine packaging (Ecovative Design LLC, 2019b)

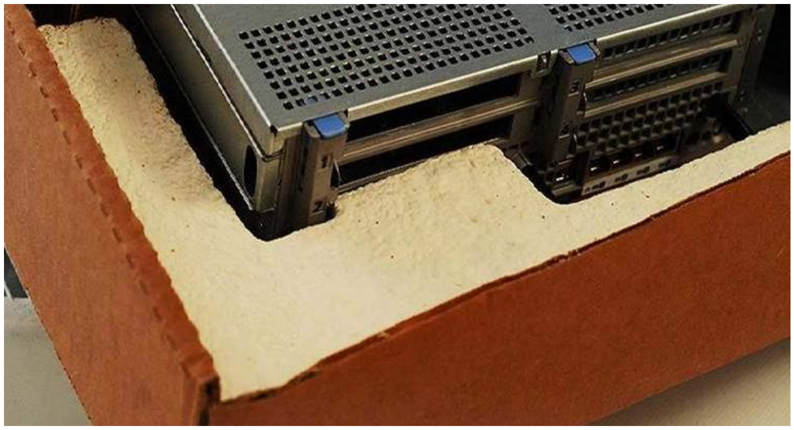

Figure 45. Ecovative Design LLC’s fungi-based materials designed for Dell electronic component packaging (Ecovative Design LLC, 2019b)

Ecovative Design LLC markets their fungi-based materials as a degradable alternative to polystyrene packaging, and whose several studies concluded that a variety of mycelium bio-composites from agricultural substrate feedstocks create 100% biodegradable materials, with high elasticity, light weight, buoyant, and flame and water resistant properties, depending on mycelium growth (Holt et al, 2014a). Mycelium materials for bulk production has so far been grown in near-net shapes, with the hyphae structures forming blocks, for such applications which require architectural forms. An example of this is the small concept building ‘Mycotectural Alpha’ created by Professor Philip G. Ross (shown in Figure 2) The East-coast USA based company Ecovative Design LLC has also focused on growing bulk biomaterials in molds, due to these shapes being easy to make and maintain during growth. Ecovative Design also supply a Material Safety Data Sheet, which established the material poses no physical or health hazards, required storage conditions (room temperature and dry), and a range of manufacturing and disposal information (MSDS, n.d.). As more commercial options for bio-based materials are created as the market for more sustainable materials grows, mycelium materials seem well placed to be developed into additional packaging and other applications such as insulation.

One of the key features mycelium materials offer is their ability to dispose of the packaging by composting, allowing the material to be broken down by bacteria, flora and other fungi found in the natural soil environment. This option reduces the environmental impact of the material, by removing the use of resources to collect, transport, and destroy materials at the end of their use phase. Ecovative Design LLC has created one of the few commercially available mycelium materials with publically available information on the cost of the material. Their packaging products range from $10.00 to $16.00 US Dollars, with $10.00 for roughly a quarter of a cubic foot volume of mycelium material, such as to package a wine bottle (Ecovative, 2019b). Similar wine bottle packaging made of cardboard and polystyrene costs $5.00 (Uline, 2019), indicating mycelium material packaging is double the cost of the current materials. While consumers may be willing to pay a higher price for a product that offers less impactful disposal options, and does not use up limited resources such as crude oil to make polystyrene, mycelium materials are still a most costly product.

The novelty of fungi as a technology is also a limiting factor. Other fungi technologies, such as brewing and bread making, have been practiced for thousands of years and have led to finding ways to mass manufacture at low costs. As packaging made of fungi grows as a niche market, it is likely innovation will be developed to reduce of cost of these materials. Compared to typical polymer packaging, mycelium materials are currently more expensive. This presents a barrier to adoption for consumers, and to large companies where the lowest cost product is often chosen irrespective of the other benefits. The global plastics packaging market is predicted to be worth $269.6 billion US Dollars by 2025, and is currently growing at a compound annual growth rate 3.9% (Grand View Research, 2018). The North American plastic packaging market for containers currently valued at $31.2 billion US Dollars in 2017 (Research and Markets, 2018). With such demand for packaging and continuous growth in the market, it suggests a good potential for mycelium materials, as relying solely on limited oil-based materials is not sustainable in the long run. If mycelium materials are currently twice as expensive as polymer foam and paper packaging, then to become completive the costs of mycelium materials need to halve. This could be achieved with economies of scale, reducing the cost of the material by producing larger volumes, and having stronger negotiating power while securing payment for waste as a feedstock. For example, if the largest retailer of wine in the USA, Costco, sold around $1.69 billion US Dollars of wine in 2015 (The Wine, 2016), roughly 70 million bottles of wine for an average $20.00 per bottle. This means an estimated 35 million pounds of plastic packaging, based on 0.5 pounds of plastic per package (Uline, 2019). If only 1% of its 35 million pounds of plastic could be replaced by mycelium packaging, this could save tons of CO_2 and millions of dollars of impact.

Conversely, the cost of plastic packaging pollution to our society is equally bad, if not worse. The financial cost of plastics to the environment has been estimated at $139 billion US Dollars per year by Trucost, a research division of Standard & Poor who specialize in providing financial information on environmental impact (The Economist, 2018). Half of this impact is from climate effects of green-house gas emissions from producing and transporting plastics. A further third is from cost of disposal, and the associated impact on our planet’s air, water and land pollution, and these impact on human health. The true cost of social harm, including the long-term impacts that are yet to be discovered is unknown, but irrespective of the financial arguments, the key reason to develop and adopt mycelium materials as alternatives to polymer packaging is to reduce harm to our fragile ecosystems. As plastics breakdown and fragment they create micro-scale pieces of plastic known as microplastics. These which have direct and indirect harmful effects on marine life, ranging from obstructing the gut of marine mammals including whales (National Geographic, 2018), to changing the composition of marine sediments and physical conditions of the marine habitats (Solomon & Palanisami, 2016). Finding alternatives to polymer materials is a key part of addressing these environmental harms, and mycelium materials can provide a biodegradable, waste digesting option to replace plastic packaging.

Out of the 322 million tons of plastic produced globally in 2015 (PlasticsEurope, 2018), plastic packaging accounted for 155 million tons (Geyer, 2017), equating to roughly 50%. Although the mix of plastic types used in packaging is not the same as for all plastics, for simplicity I'll assume the externality per ton is the same. Externality here is defined as the costs of negative impacts of the plastics on the environment, public health, and wider society. Based on this, packaging is then 50% of the approximate $139 billion US Dollars externality plastics cause. So the externality per ton of plastic packaging is estimated as $450 US Dollars externality per ton of plastic packaging.

It is important to note that these calculations hide many assumptions, and ignore the enormous variation in the price and social cost of different plastics. So it is likely some plastic packaging has a higher social cost per ton and as a share of price (and others are lower). So socially or environmentally conscious consumers who care about consequences should be willing to pay a premium for mycelium materials. Alternatively, efficient (Pigouvian) taxes on plastics (which are taxes intended to punitively correct the negative outcomes of using plastics), would make mycelium materials more cost-competitive. This would require the mycelium materials to be grown at a greater scale, and the costs of designing, producing and transporting standard shipping products would need to be less than this externality cost. Additionally, this also requires a mycelium material packaging that is 100% comprised of natural, biodegradable materials, while still meeting the strength and weight performance requirements for packaging.

Case Study Two: Biomimetic Fabrics

The need for materials with lower environmental impact during manufacture and disposal is a key area of development for biomaterials. An area for innovation is materials which improve the sustainability of consumer products. In particular there is an increasing trend for improving the sustainability of products which users have close contact with, such as textiles.

Consumer research also shows that eco-materials are becoming a fast expanding area of concern and innovation within the textile and clothing industry (Niinimäki & L. Hai, 2011). Consumer textiles and apparel have relatively short use phases, require fast manufacturing times, and have limited recycling and reuse options. While there is a long history of natural materials used for textiles and clothes, such as cotton fabrics, production of natural fiber materials is not without impact, including the use of fertilizers and chemicals during growth and manufacturing of the actual product (Blackburn, 2009). This environmental harm makes developing biomaterials that can be used as alternatives an area ripe for innovation. While there have been significant efforts to create solutions, including the increasing use of organic fibers and environmentally safe dyes (Gam & Banning, 2011), development of biomaterials to offer alternatives for the more durable and tough materials such as leather, has yet to be a major area of focus.

Synthetic composite laminates are formed of layers of polymeric material combined with layers of fiber reinforcement, such as carbon fiber epoxy laminates. These laminates are typically high strength and high performance materials. Their relatively high strength comes from their layered structure, with the lower-strength polymeric matrix being reinforced with the fiber layers (Horrocks & Anand, 2000). Composites combine the properties of their constituents, creating a composite material with the combined useful properties of both materials. The resulting desired properties, such as toughness and stiffness, can be designed by materials selection and combining materials with these properties. Common synthetic matrix materials are thermoplastic and thermoset polymers, and common synthetic reinforcements are glass, aramid and carbon fibers in various fiber roving forms, such as random fiber and woven mats.

For low-stiffness applications, such as for flexible materials, composite laminates can also be formed into flexible fabrics. These fabrics are typically used for light-weight architectural applications, such as the roof fabric of the McClain Center at the University of Wisconsin, with a flexible composite formed of two layers of polytetrafluoroethylene glass fiber fabric with a core of translucent aerogel (Wright, 2015), making a light, flexible and optically translucent material.

Biocomposites share many of the characteristics of synthetic composites, again being formed of a combination of a matrix and a reinforcing materials, except some or all of the materials are biological in nature. Due to the relatively heavy environmental impact of synthetic composites, biocomposites are continuing to be developed by academics and entrepreneurs to help make them less impactful, and more widely available (Mohanty et al., 2002). Materials partially made from biomaterials, such as jute fiber reinforced with synthetic epoxy composites, are an improvement in terms of environmental impact, but are still not entirely recyclable or able to be disposed of using natural methods such as composting, due to their partially synthetic and environmentally damaging components. Biomaterials composites usually require two different types of materials, such as a bio-derived polymer matrix, such as an environmentally degradable poly-vinyl alcohol (Chiellini et al., 2004) and a natural fiber reinforcement such as cotton.

Materials made entirely from natural substances can provide the advantage of being able to be disposed of via composting, and due to not being formed from crude-oil derived substances, can have reduced environmental impact in their manufacturing phase. Mycelium composites can provide innovations to help meet these needs, as they are formed entirely from natural materials. They can be formed into biocomposite laminates, made of the mycelium material as a matrix, and a natural fiber reinforcing laminate inside of the matrix. Similar to synthetic composites, mycelium materials can be designed to meet specific needs by selecting biomaterials to combine as a matrix and reinforcement.

Fungi-based substances can be used to develop materials that can offer the sustainability and disposal phase impact reduction that is desirable from bio-based composite materials. Due to the branching, space filling growth mechanisms of fungi (Edelstein & Segel, 1983), mycelium materials can be grown in a large variety of forms, by modifying the growing conditions such as temperature, light, humidity, and gas levels (Chau, 2016). The development of mycelium materials in flat sheets, with the hyphae forming layers of mycelial growth, effectively forms a laminate structure. This innovation was developed by the west-coast USA based company MycoWorks. Due to the tough, pliable properties of the mycelium-based material, it forms a flexible laminate similar to textile fabrics and animal hide materials. While the growth conditions and manufacturing methods are still being developed, and remain largely the private intellectual property of the developers, basic manufacturing methods involve selection of a species of fungi, providing a feedstock, physically controlling growth of the mycelial masses, and denaturing of the grown material.

Biomimicry is a method to solve problems in a way inspired by nature, with the term literally translating as ‘life-copying’ (Lurie-Luke, 2014). Due to the constant competition and constraints on resources in the natural world, nature designs solutions to challenges in a highly efficient and low-impact way, providing a rich source of inspiration for engineering problems. By mimicking the way nature solves the problem, solutions can be found to design challenges which traditional design methodologies struggle to solve efficiently. Biomimetic solutions need not just be copies of nature, they can also mimic the design processes and strategies nature has developed.

Biomimicry can also provide insight as how to solve complex material demands that seem contradictory, in a highly efficient way. Similarly, mycelium materials can solve seemingly contradictory material properties, such as being flexible and tough, while still being made of natural materials and not being animal derived. Due to mycelium materials' naturally occurring structure and composition of chitin, the material can be grown in laminar structures which mimic the layered structure of animal hide, having a composite structure made of layers with distinct morphologies. Part of this laminate structure is created by the natural formation of mycelium materials hyphae, which grow into a cellular material matrix with reinforced areas rich in chitin. By selecting mycelium materials to mimic the morphology of a natural material, such as leather, replacement materials for animal hides can be created. These materials can have similar surface textures and material properties such as flexibility, toughness, and compressibility, and similar visual appearance to leathers, as seen in Figure 46. Post-processing of the resulting laminate mycelium materials may be required to form the desired surface finishes, and also create the resulting degree of flexibility and pliability of the final material. These material properties can be achieved by both mechanical and chemical processes, although the entirely natural composition of mycelium materials means only natural processing substances are preferable.

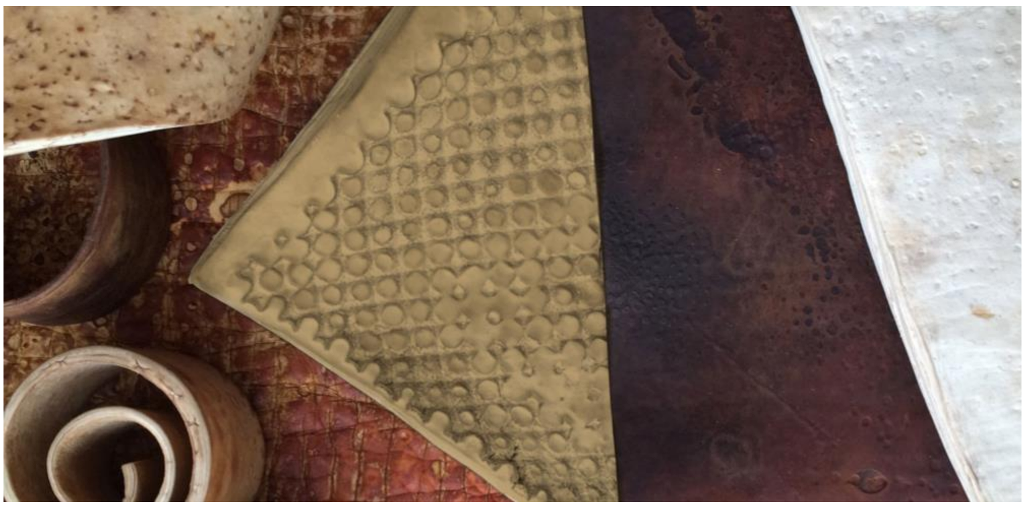

Figure 46. Various mycelium laminate materials as alternatives to leather materials created by MycoWorks (MycoWorks, 2016)

The use of mycelium materials to replace other natural materials such as leather seems a surprising use of biomimetics. The driving motivation is the ability of mycelium materials to provide a lower environmental manufacturing impact and to provide a low-impact disposal phase by composting. While reducing the impact of synthetic materials has focused on their reuse or recycling into other products, overall this is still a costly exercise in recovery, storage, reprocessing, and one the heavier impact phases of recycling, transportation of the materials. Consumers also continue to drive the production of first-use, non-recycled products, particularly for fiber products that have close contact with the user's body, such as apparel.

While one would think all natural materials have a lessor environmental impact than synthetic materials, this is not always the case. Many of the processes used in the production of natural materials made from animal products are highly toxic and (in many nations) heavily regulated. These regulations make these materials more costly to produce.

A market with potential to benefit from innovation is animal-based leathers. Animal leathers require lengthy processing, including preparatory soaking, mechanical processes such as de-fleshing, splitting of the layers of animal skin, and multiple chemical processes including pickling and degreasing, and liming. Liming is a key step, as the alkali solution denatures and removes inter-fibrillary proteins, which form the protein connective and supportive structure of the leather, which enables the material to become flexible and tough. These processes are environmentally damaging. Tanning processes generate noxious hydrogen sulfide gas and toxic solid wastes, including lime and chrome (Thanikaivelan et al., 2004). Historically tanning was such an unpleasant industry that leather processing was relegated from urban areas. Some leathers continue to be produced in low-resource countries where regulation is less stringent than in highly developed countries, resulting in a largely unknown environmental impact, and risk to workers and people living near the processing sites, particularly from cancer (Decouple, 1979).

The market for non-animal materials also continues to increase due to the rising popularity of non-animal product-free lifestyles such as various disciplines of veganism. Veganism, the key feature of which is the commitment to non-use of animal-based and derived products (Cherry, 2006), is a growing cultural movement, and consequently a growing market for products.

Due to the increasing awareness of consumers of the resources used to produce their products, knowledge of externalities, high costs of regulation and the goals of vegans, non-animal based leathers are gaining acceptance. While these wider trends of responsible production and 'eco-friendly' consumer buying habits are still growing, their impact on the development and creation of new non-animal based products is becoming more visible. Luxury trend-setters such as Stella McCartney are highlighting their use (L.A. Times, 2015), bringing more attention to the market for non-animal and non-synthetic materials that helps feed a slowly growing market (Panteva, 2012). One barrier to overcome in the production of leathers from non-animal sources is the issue of scaling current production methods. Current production of vegan leathers are decentralized and mostly small-to micro scale businesses where production is an individual process, akin to cottage industries (Payne et al., 2016). For wider use of non-animal and non-synthetic leathers, including those made from fungi, production needs to increase as does the volumes of materials being produced, to help provide a constant supply chain which product manufacturers can rely on. Some advances have been made in culturing materials from cells (Ščerbaka & Ulme, 2016). Unfortunately, the process of culturing animal cells and forming them into layers of material is costly and time intensive (Freshney, 1986).

Due to fungi being able to digest a wide variety of organic substances, from agricultural wastes to crude oil-based synthetic materials, mycelium materials also have the potential to be an alternative to animal-based hides such as bovine leather. A material currently being developed by MycoWorks as a replacement for leather in shown in Figure 47. Mycelium materials can offer a lower-impact potential alternative to leather with a more ecologically positive disposal option via composting.

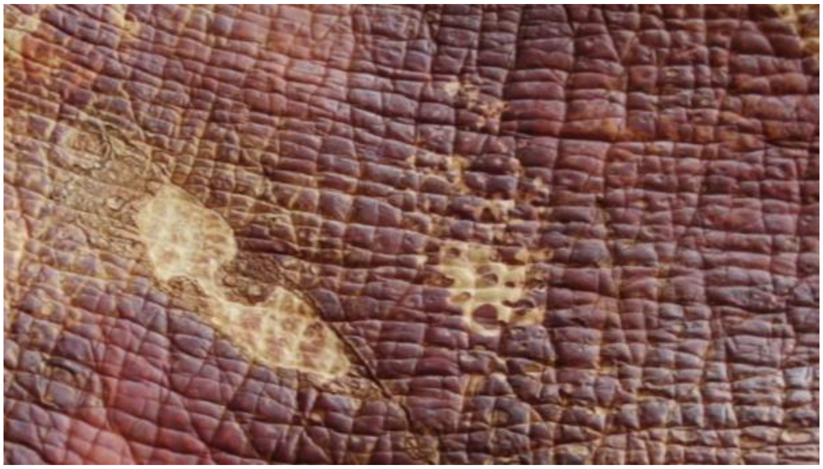

Figure 47. Fungi-based leather created by MycoWorks (Chau, 2016)

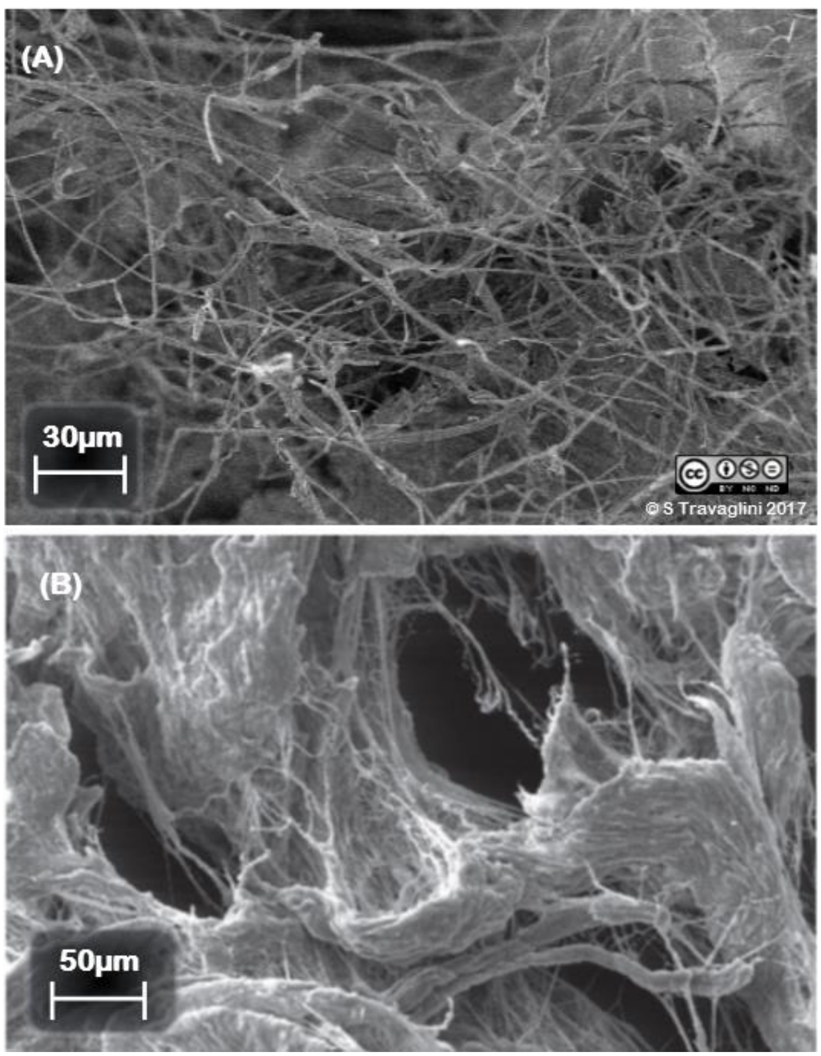

Figure 48. Scanning electron microscopy images of materials microstructure
of (A) mycelium materials, and (B) shaved leather (leather image adapted from Ribeiro et al, 2012)

To compare the microstructure of laminate mycelium materials and the microstructure of animal hide leather, scanning electron microscopy (SEM) was competed on the mycelium material. Images of the microstructure of mycelium materials in Figure 48 (A), and of shaved animal leather in Figure 48 (B), exhibit similarities in the reinforcing, fibrous tangle of mycelium material's hyphae and leather's fibrous proteins. Animal leather hides are made of layers of protein structures, such as the uppermost layer, the grain, which forms the visually recognizable surface of leather (Michel, 2014). Within these layers, fibers form the structure of the leather, and can be seen in Figure 48 (B) forming tangles and individual stands of fibers. These give the leather its toughness (Liu et al., 2009). The layers of the mycelium materials show similar structures, with fibers forming tangled masses, with layers of chitinous fibers in various densities. This may provide clues to why fungi-based materials can form close replications of animal hide characteristics and morphologies, despite being made from significantly different protein structures. The mycelium materials shown in Figure 46 show their flexibility and thickness similar to animal leathers, and how the mycelium materials natural surface texture is similar to animal leather in Figure 47.

The ability of mycelium materials to fill a mold volume during growth, and to naturally grow in variety of textures, lends them to further development. For example, they can be grown in mold to take on the texture of rare leathers, such as crocodile leather. Mycelium materials can offer several advantages including being able to be grown from waste feedstocks, faster growing times, and less resource intensity than animal-based materials. The recent innovation in the field of fungi-based materials, of textiles grown in layers to form composite laminates (Chau, 2016) is a potential replacement for animal-based hides such as bovine leather. With both the matrix and reinforcing fibrous structures made of naturally occurring chitin, and fungi's ability to digest a variety of feedstocks dependent on the selected species of fungi, mycelium materials can offer an alternative to synthetic and animal-based textiles and can offer a less resource intensive biomaterials option. Overall, innovations in fungi-based biomaterials can offer new directions in biomimicry, including developing alternatives to animal-based materials using laminate mycelium materials.

Chapter Eleven: Future Applications

Due to the novelty of mycelium materials, many of the applications they are suited to are yet to be developed and proved as a viable technology. Instead, some potential applications are currently conceptual, and the following case study has been conducted with the aim of inspiring further work in the area of using fungi-based materials as a sanitation technology.

Case Study Three: Remediation & Sanitation

The abundance of fungi in every known environment (Ranzoni, 1968), and the need to find sanitation solutions for human waste that do not pollute the environment or require costly, urban infrastructure to be effective (Austin & Van Vuuren, 2001), is the motivation for developing mycelium materials as a sanitation technology. With approximately 800,000 deaths per year in low- and middle-income countries, due to inadequate sanitation, drinking water & hand hygiene (Jannet et al., 2018), new technologies which can be efficacious in low-resource situations is a potential application for mycelium materials and the wider fungi family.

The main idea being explored is using fungi as a sanitation technology, as both a remediation technology as it currently stands, and ideally for use in a composting toilet that can produce a useful or valuable material as its output. As using fungi to repurpose human waste is a novel topic, the closest technologies were considered, such as environmental remediation. This includes a brief literature review to introduce the key information needed to understand fungi's potential role as a remediation and sanitation technology.

To understand the sanitation applications of fungi technologies, first the abilities of fungi to remediate environmentally damaging wastes need to be described. Specific species also need to be identified as potential starting points for the development of such technologies. Fungi have developed alongside plant and animal development, with the majority of fungi living in close association with other organisms, chiefly plants, to get their required nutrients. Their development over time has also depended on the development of the ecosystem and our impact on it (Gladieux, 2010). Their highly-attuned design to a wide variety of specific environments results in being suited to equally specific feedstock, ideal growing conditions, and the resulting fungal physical structure and form. When these align with the purpose and environment we are trying to impact, or design a product for, new opportunities for using fungi in a variety of applications can be developed. Using fungi for remediation is a well-established tool in the field of environmental control and restoration, due to their ability to digest an incredible range of organic and in-organic matter, depending on the species' preferred feedstock and environment. Thus far fungi have been successfully used for remediation in a variety of situations, due to their ability to:

- Uptake and safely contain heavy metal contamination (Galli et al, 1994), Khan et al., 2000)
- Remediate oil-containing soil (Yateem et al., 1998)
- Remediate waste substances from beverage processing such as distillery wastewater (FitzGibbon et al., 1998).

The mechanisms of fungi remediating pollutants is an extension of their feeding and growth mechanisms, using enzymes to break down matter and convert it into useful substances. The fabric dye industry has been a forerunner of using fungi for remediation, due to the large volumes of polluted water produced and their resistance to microbial degradation. (Novotný et al., 2004) used *Phanerochaete chrysosporium, Trametes versicolor, Pleurotus ostreatu*s and *Irpex lacteus* to degrade recalcitrant compounds, which are xenobiotic synthetic chemicals that are extremely resistant to degradation in the environment by natural organisms (Fernando & Aust, 1994). Novotný et al. achieved the breakdown of these substances in dye byproducts through the fungi's production of Mn-dependent Peroxidase (MnP), and Lignin Peroxidase and Laccase (LAC), which can breakdown dye byproduct pollutants through enzyme action (Novotný et al., 2004). FitzGibbon et al. (1995) researched the use of various fungi to remediate molasses spent wash concentration, a waste byproduct from beverage distillation processes. It contains a high pollution load due to its color and potential for Chemical Oxygen Demand (COD), a measure of how much oxygen will be used as the spent wash decomposes. The spent wash also contains phenolic compounds that inhibit microbial action (Borneman, 1986). The color of the spent wash, phenolic compounds, and having a high COD contribute to causing eutrophication and harm to the environment. The use of white-rot fungi, specifically *Phanerochaete chrysosporium* and *Coriolus versicolor* were found to remediate the molasses spent wash concentration giving an overall reduction in COD by 77%. Wesenberg et al. (2003) investigated white-rot fungi such as *Phanerochaete chrysosporium* and *Trametes versicolor* etc., for the decolorization and detoxification of industrial dye effluents, though the action of lignin-modifying enzymes. The most efficient removal of the pollutants was achieved in aqueous solution, rather than in a soil substrate, which suggests fungi species suited to arid environments would be a key requirement for any remediation system used in counties with low water resources.

Fungi are a key part of all ecosystems including in arid conditions, with research in desert environments showing the presence of fungi in three desert areas in Namibia associated with animal communities (Ciccarone & Rambelli, 2000), and (Ciccarone & Rambelli, 1998). Thermophilic fungi can thrive in temperatures up to 62°C (143°F) (Maheshwari et al, 2000). Thermophilic fungi have long been studied in relation to their role in composting, with research showing they are central to the decomposition of waste, including municipal waste (Kane & Mullins, 1973), with a key requirement being their aerobic respiration. The above examples suggest that through careful species section, fungi could be used to remediate environments contaminated with organic and in-organic contaminants, in low-resource conditions such as arid environments with a lack of sanitation facilities. Ligninolytic fungi have been found to degrade environmental pollutants such as munitions waste, pesticides, organochlorines, polychlorinated biphenyls (PCBs), Polycyclic Aromatic Hydrocarbons (PAHs), synthetic dyes, wood preservatives and also synthetic polymers (Pointing, 2001). There are even fungi which have the ability to exploit radiation from nuclear waste, in a process known as radiotropism. This is the ability to harvest usable energy from ionizing radiation, such as gamma radiation emitted from nuclear reactors (Dadachova & Casadevall, 2008). Some fungi have also been found to seek out radiation at the Chernobyl disaster site (Zhdanova et al., 1991), as exposure to radiation enhanced the fungi growth. While there is little information on how much energy fungi are absorbing during radiotropism, it does suggest that fungi could be developed as a technology to produce useful materials in nuclear disaster areas.

Fungi's ability to digest carbon-based materials including polymers, has already led researchers to identify several species that digest waste polymeric materials. Yamada-Onodera et al, (2001), found *Penicillium simplicissimum* can digest and degrade polyethylene, and enzymes produced by *Aspergillus flavus* degrade the mechanical performance of polyethylene bags (El-Shafei, 1998). The speed of polymer degradation by fungi is fast when compared to the natural degradation rate of polymers, which takes hundreds of years. Prashanth & Tharanathan (2005) found polymethyl methacrylate (PMMA) film was degraded after 25 days of exposure to *Aspergillus flavus*, and polyamide fibers being degraded by *Phanerochaete chrysosporium*, reducing the molar weight by 50% in 90 days showing the fungi were attaching and breaking the hydrocarbon chains in the polyamide, effectively dismantling the polymer. Bredberg et al. (2002), reported *Resinicium bicolor* being effective in degrading waste rubber tire products. The ability of fungi to remediate toxic and hazardous substances (including inorganic ones), without creating yet more by-products, is unique. It lends the development of fungi technologies to environments where multiple types of remediation is needed, which fungi could achieve through organic means without needing costly or labor intensive resources.

Mixed sources of waste could also be addressed using fungi as the degradation mechanism, as while the majority of problem substances that remediation aims to degrade are organic in nature, in-organic substances can also be part of fecal and waste matter. This could be due to sorting of waste not being possible, which could be addressed by fungi species which can digest organic and in-organic matter. This concept is a research topic in its own right, and if the resources were available, further research could be completed into which species digest various materials via a literature review in the next section. The interaction of these species, and ability to effectively digest waste also needs to be investigated via empirical testing. Lastly, the safety of introducing non-naïve fungi to an environment should be considered, and native fungi species used where possible to reduce the risk of invasive fungi species damaging an environment. Considering the explored abilities of fungi to digest and remediate a range of environmentally harmful wastes, fungi technologies could provide a new range of waste technologies. They could drastically reduce the time needed to degrade polymers, and could also suggest a way to address the vast waste landfill sites already in existence, that contain millions on tons of plastic that will otherwise will not naturally degrade within a meaningful human life time.

Coprophilous Fungi for Sanitation

A whole subset of the fungi kingdom, coprophilous fungi, specialize in fecal matter as their preferred, and sometimes exclusive, source of nutrition. Many species of coprophilous fungi also grow directly in feces, having a life cycles that work within digestive systems and only bloom (via the production of spore-delivering mushrooms) once exposed to the outside environment. These could suggest a vast resource of fungi well suited to addressing sanitation needs in a variety of environmental conditions. Since coprophilous fungi are suited to digesting fecal waste, and can convert it into useful materials made from their mycelium, there is the potential to create a composting toilet based on fungi. This concept is motivated by the wider need for sanitation technologies that can reduce environmental impact, and also to create an opportunity to make useful by-products from human waste in locations with few resources. The potential to create a waste containment system that is entirely made from natural materials is a key advantage for reducing the impact of the toilet, both in terms of safely disposing of the human waste, and also not using oil-based materials for the physical waste container. Various use cases for composting toilets include:

- Low-resource residential locations which lack infrastructure, such as slums
- Short-term temporary housing such as refugee camps
- Dispersed campgrounds or remote public facilities

The fungi technology could be used to make the entire toilet system, including the pipes, privacy enclosures etc., or just the waste container. Two possible cases are considered for processing the waste, after the final use of the toilet when it is full. The waste container could be buried, with the fungi facilitating remediation underground. Or the waste container, pre-seeded with fungi designed to make functional products from the waste, could be removed and used to form useful products. An overview of a possible process is shown in Figure 49.

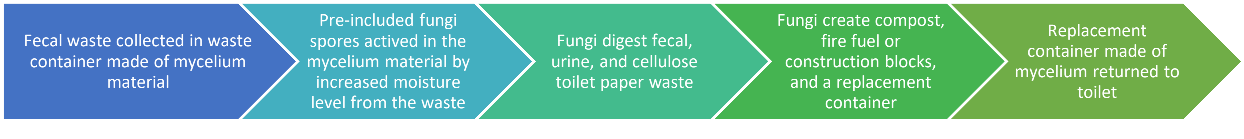

Figure 49. Mycelium container-based process for fungi-based sanitation application

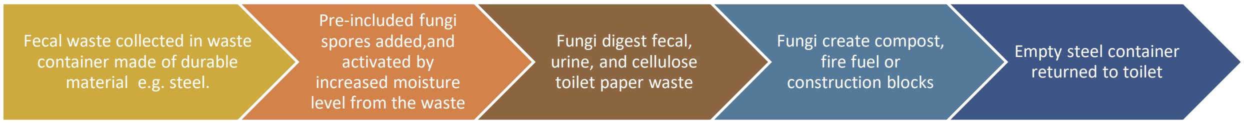

Figure 50. Durable container-based potential process for fungi-based sanitation application

In the first case, shown in Figure 49, only the waste container that collects feces would be removed from the toilet (such as a bucket-shaped container made of mycelium material under the toilet seat), and then buried. This means the toilet could be located a close but safe distance from homes, increasing the likelihood that users would choose to use the toilet. This is important as providing toilet facilities in low-resource locations, does not always result in toilet use – barriers to use included preferring open-defecation, preferring privacy/toilet enclosure, and toilets being in a convenient location close to homes (Sinha et al., 2017). A possibility to address this issue could be to include waste containers made of mycelium materials designed to be located in location typically used for open-defecation, which could then be buried after use to prevent the spread of pathogens from the waste, or waste run-off. Fungi have fast growth rates that could remediate the waste and run-off faster than the natural decomposition of waste either above or below ground.

In the second case, shown in Figure 50, the waste container could be removed from the toilet, and the fungi allowed to digest the waste and turn it into useful products. Since fungi species exist that specialize in digesting waste, and other species which grow useful materials or edible food from various feedstocks (including human waste, and other fungi themselves), these could be combined to create a set of fungi that first digest human waste and then create a useful material or resource through digesting the waste. This could be achieved through breeding fungi or genetic manipulation to create a fungi species that digests human waste, and grows useful mycelium materials or edible food. It is important to note that this is currently an untested concept, and more development would be needed to determine if edible mushrooms grown from human waste are safe for human consumption, the time limits needed to achieve this, and user attitudes to such a technology. While it is possible users may refuse to consume an entirely safe food source originating from a human waste, livestock animals such as goats and cows have no such issues. This opens the potential for fungi to grow food for livestock from human waste toilets with fungi technologies.

While the effectiveness of fungi to digest human waste is yet to be studied in an academic setting, Paul Stamets, the noted fungi expert and long-standing proponent of environmental remediation using fungi, recently reported that fungi are up to 100% effective in removing bacteria harmful to human health from storm water (Taylor et al., 2015). Stamets termed this process 'mycofiltration'. One of the pathogens from human waste and fecal matter that evidences contamination of water is Escherichia coli (E. Coli) (Centers for Disease Control and Prevention, 2018). Past success by Taylor et al., and Stamets in removing E Coli from contaminated soil using fungi is an example for future research efforts to follow, when considering how to remove pathogens associated with human waste. Due to this success, E Coli was selected as the area of focus to consider fungi's ability to make human waste safe, although there are other harmful pathogens which would also need to be addressed by fungi used to make matter from a composting toilet safe. The efficiency of the process to remove bacteria, in Taylors' research (2015) E. Coli suspended in storm water, is chiefly dependent on the fungi species selected, the processing system used to filter the water, and many factors yet to be fully identified or shared in the public domain.

A key factor of the fungi-based filtration systems is the reliance on fungi that have the ability to not be impacted by excessive heat, cold, saturation or dehydration. Due to the millions of species of fungi (Hawksworth, 1991) and the fact they specialize in every known environment, it is likely to be possible to find a naturally occurring or genetically creating a fungi species that is suitable to remediate human waste, with the ability to remove a specific pathogen. In addition, as specific locations and ecosystems naturally develop and support specific fungi that thrive in that environment, the presence of a particular food source (including pathogens) is often connected to appropriate fungi co-existing there. The growing ability of mycologists to identify, select and adapt various fungi, through selective breeding or genetic manipulation, means there is an opportunity for fungi to be designed to remediate areas contaminated with human waste, or directly digest human waste. This conclusion was also reported in the research by Taylor (2015). Taylor reported the suitability of various fungal strains to be effective in the removal of pathogens from water, but also acknowledged the need for further research to find the species and conditions to safely process waste as an industrial process.

In settings where water is an important medium through which soil contamination hazardous to human health occurs, such as for the soil around a latrine, human pathogenic fungi could also be transported by run-off water. While the number of fungi present in water is extensive with Mara & Horan (2003) noting a non-exhaustive typical list of 46 fungi species, including many *Aspergillus, Zygomycetes, Alternaria, Cladosporium, and Phoma* species (Mara & Horan, 2003). Some of these pathogenic fungi are themselves considered potential secondary pathogens, meaning pathogens which are not primary causes of disease in healthy adults, but can cause harm when the human immune system is compromised. It is key to note that locations in need of latrine sanitation solutions often have low resources and higher rates of immunocompromised individuals and children, due to the lack of sanitation, and thus likely to have high concentrations of pathogenic fungi in water (Cooke & Kabler, 1955). The solution may not in fact be the eradication of fungi in the water as part of a sterilization process, but the provision of an environment that allows the domination by a single fungi species, or set of species, that prevents the pathogenic fungi being effective or able to act as disease vectors.

Another option could be for fungi to be used to produce pathogen-killing substances, creating a way to naturally sanitize pathogens from an easily-shipped starter culture of fungi spores, which would be a useful technology for areas with limited resources, sanitation infrastructure, or transportation links. *Muscodor albus* produces antimicrobials in the form of bioactive volatile organic compounds that interact to denature a wide variety of plant and human pathogenic fungi and bacteria (Strobel, 2006). Various fungi species can also produce hydrogen peroxide H_2O_2 which is a bleaching agent and antiseptic, suggesting a potential way to sterilize matter via fungi (Koenigs, 1974). Fungi also exist that are specialized to grow in conditions high in urea or urine, with Lehmann (1976) identifying species that thrived in cellulosic-based pinewood matter soaked in urine, including several species that only thrived when urine was added, including *Ascobolus denudatus*, *Coprinus echinosporus*, *Tephrocybe tesquorum* and *Pseudombrophila deerata*. This suggests human waste could be separated, such as in urine-diverting compositing toilets, and fungi specializing in solid waste and urine separately used to either remediate or speed up the degradation process of a compositing toilet.

Lastly, the secondary purpose of fungi sanitation technologies after waste remediation, would be to produce a useful, safe, and preferably commercially valuable by-product. For example similar technologies have been created to make useful by-products from urine, such as Sanivation's 'Blue Box', which has a urine diverting system that produces fertilizer. Exploring Sanivation's development process, and seeking their input on topics such as waste regulations, toilet design, and product development strategies, would help inform the development of fungi sanitation technologies. The following section explores the key steps needed to develop such technologies based on coprophilous fungi.

Sanitation Technology Development
Since nature has already provided a range of fungi that literally live to digest fecal waste, coprophilous fungi present an excellent starting point to be developed into a technology that can digest human or animal waste and produce a useful product as part of it or as a by-product. A summary of such a process is outlines in Figure 51. One major drawback of current composting toilets is their inability to deal with mixed wastes such as plastic bags, paper products, or other common rubbish. Since various fungi species can digest feces, polymers, cellulose, and toxic substances such as heavy metals, there is the potential for a composting toilet that includes multiple types of fungi to break down such materials.

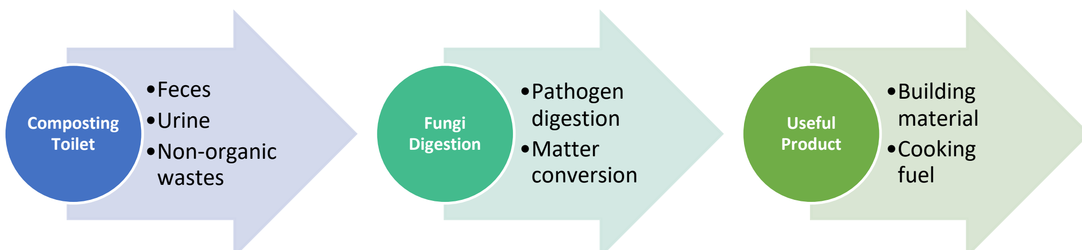

Figure 51. Potential fungi-based composting toilet process flow

The target locations for a toilet that uses fungi would be low-resource areas where there is a lack of adequate toilet facilities, such as unauthorized refugee camps, emergency situations, and rural villages without government sanitation infrastructure. The target users are members of these under-served populations, and include all genders and ages. The theory of change is that having access to a fungi-toilet would reduce rates of infection from human-waste tainted water, by reducing the transmission of pathogens from ineffective toilets, and improve rates of use by providing a useful material by-product useful to the user. Potential useful products could be:

- Fertilizer – using mycorrhizal fungi which fix nitrogen (Barea et al., 1992).
- Housing materials – this shows great promise for use with animal dung, a traditional and still widely used building material (Hall & Kenward, 1998), (Yalley & Manu, 2013), overcoming the lack of resistance to moisture by the inclusion of mycelium as a binder.
- Fuel – animal dung is used as cooking fuel in many low-resource environments, (Mekonnen & Köhlin, 2008), and fungi could be used to digest dung (or feces) and once desiccated using sun-drying, be used as a fuel. This may only be viable, if made safe in

conjunction with cook stoves developed to address improper burning and fumes (Levine, & Cotterman, 2012).

Each of these topics discussed above further analysis and validation at each stage, to determine their feasibility and development requirements. Many pathogens exist in feces, including salmonella and ringworms. While fungi are known to effectively reduce E Coli as soil remediation, and the predatory *Orbilia* species of fungi which prey on Roundworms (Barron, 2003), there are many development steps needed to ensure fungi could eliminate all the pathogens in human waste. There are a great range of development needs to design, prototype and commercialize fungi as a sanitation technology, which go beyond the scope and resources of this case study. This study does, however, highlight valuable questions that need to be explored to develop and validate it.

More investigation is needed into several aspects of a fungi-based composting toilet, including:

- **Rate of waste decomposition**
  To prevent health hazards, or excessive odor (and thus a slow adoption of the technology), the fungi would have to decompose the waste fast enough to prevent contamination of the surrounding area. The rate of decomposition in the target environment should be determined, including during various seasonal conditions and temperatures which may occur. This is also important to ensure timely and safe decomposition of the waste.
- **Degree of waste decomposition**
  Incomplete digestion of waste could pose a health or ecological hazard, or could hinder the action of other organisms such as naturally present bacteria that break down waste. This problem could be solved by selecting fungi species that don't hinder other naturally occurring waste decomposition processes, potentially by sourcing fungi present at waste-processing sites.
- **Efficiency of fungi to digest and sanitize pathogens from human waste**
  While the fungi species suited to digesting animal dung have been well-researched, fungi that digest human waste is not well researched. Ideally, testing could be carried out to determine which fungi species thrive on human waste, and their ability to safety digest or remove pathogens. It should also be determined if the diversion of urine is necessary to prevent anaerobic respiration, or if various fungi could co-exist to remediate feces and urine in the same container. For example, yeast, a member for the fungi kingdom, can grow through anaerobic respiration (Tustanoff & Bartley, 1964), and could be used to digest waste in the anaerobic conditions made by urine.
- **Efficacy of fungi technologies at digesting waste in low-resource environments**
  The technology could be impacted by user habits, for example the use of toilet paper, or the use of the toilet for mixed waste disposal like plastics, due to the lack of waste disposal facilities in low-resource environments. A solution could be to include a mix of fungi species (which again, would require investigation to determine if and how they would co-exist) that digest human waste, and paper, plastics etc.
- **Evaluation of the environmental and social benefits**
  The benefits and costs to the environment and target users should be assessed to determine if it would provide a better solution than their current sanitation options. The environmental and social impact assessment should include assessing the degree of environmental remediation, reduction in the presence or quantity of harmful pathogens, and result on health levels and satisfaction of the users. If the toilet was developed to a working prototype

stage, and production had reached a sufficient scale to deploy in several locations, impact analysis methods from public health could be used for an impact assessment. This would involve evaluating two groups of people in a low resource area, where one group is provided with fungi toilets, and the other is not. The disease rates of the two groups could be compared, such as using statistical testing of independent data about the incidence of illness in both groups relating to lack of proper toilet facilities, to determine if the technology does result in improved sanitation conditions and health outcomes.

- **Determining potential cost per toilet**
  For a fungi toilet to be a viable project, the costs of manufacturing, transporting and maintaining a fungi-based toilet also need to be determined. The design and continued costs implementing a fungi toilet should be carefully assessed and tested, to prevent the costs of developing the technology exceeding the likely development funding, or the on-site costs exceeding what the target users are likely to be willing to pay.
- **Validating the usefulness of by-products**
  If the fungi toilet is used to create useful by-products, such as fertilizer, construction bricks, of fuel for fires, the efficacy of creating these and their usefulness should be verified with testing and prototyping. For example, if used as a fertilizer, the ability of the material to improve the soil condition and crop outcomes must be validated and evaluated.
- **Exploring user needs and adoption behaviors**
  Prototyping a compositing toilet is key to discovering the design and function requirements through user testing. More knowledge is also needed to understand how to learn about the target users, their needs, their cultural values around sanitation, and their likelihood to adopt fungi-based sanitation technologies.

One source of information would be to collaborate with entities connected to UC Berkeley with experience in sanitation technologies for low resource areas, such as the Sanivation and Sanergy. Collaborating could help find key information needed to develop fungi-based toilets, such as the regulations and laws controlling fecal technologies, research and testing methods for the design, manufacturing and safety of such toilets, and also methods for user research to understand user needs and choices in toilets and sanitation.

Finding the fungi species to digest waste and safely remediate pathogens found in feces and urine, and potentially producing a useful product from the fungi material itself is a complex task. Literature reviews and contacting fungi experts could be used to identify candidate fungi, along with connecting to local mycology enthusiasts, who often have a deep wealth of knowledge of native fungi and their preferred food and environment. One potential avenue of investigation would be to collect and select fungi already suited to digesting waste, such as those found at municipal waste treatment plants. Another would be to explore the fungi species present in low-resource locations, such as the locations of human waste disposal in slums. This will determine which fungi species are suited to thriving in a high urea, high fecal matter environments. The ability of these species to thrive on human waste would also have to be tested, measuring their outputs, time taken to grow, optimal growing conditions etc.

Another approach could be to genetically manipulate or breed fungi species specifically suited to digest human waste. There are many coprophilous species that specialize in digesting fecal matter, or thriving in high-urea environments (Lehmann, 1976), (Mara & Horan, 2003), that could be combined or bred to be used in such environments.

Different fungi species may be required to breakdown the waste, and subsequently create a useful outputs, at different times between the creation of the waste and the end of the entire process. As fungi may compete or fail to thrive if multiple species are growing at the same time. This could be solved by introducing each fungi species at a specific time, using time-delayed capsules. A novel concept created during this research could be to design and apply a time-delayed container for fungi spores, which breaks down during or after the use period of the toilet, allowing selected species of fungi to colonize the waste at a specifically chosen time. This time-delayed feature could be achieved by coating a plug of fungal spores in a cellulose capsule that degrades over a specified time, allowing the fungi to digest the waste, or even digest different species of fungi already present/introduced to the waste. This could be leveraged to use one (or a set of species) of fungi species to break down and sanitize the waste, and then another species (or set) to digest the original fungi and turn them into useful materials or products. For example, for the fungi-based container that is buried when full of waste, fungi spores could be contained in a capsule embedded in the container that aids the safe and timely digestion of the waste, while also adding fungi that benefit the soil. For the reusable container, capsules of dry inactive fungi that turn feces and urine into building blocks for homes could be added to the container, which activate and grow due to the wet conditions.

Testing the efficiency of fungi at digesting and sanitizing feces could be established empirically, by assessing the ability of fungi species to reduce the number or effectiveness of various pathogens found in human waste in a laboratory setting. Further work is needed to investigate the range of pathogens in human waste, which fungi species can digest or inactivate these, or which species could create an environment where such pathogens cannot exist. Testing could initially focus on the species identified by literature review, and then expand to species selected from the environment the composting toilet would be used in. Further work could also be completed into including bacteria and other biological vectors that remediate human waste, and how these affect the growth of the fungi species, and efficacy of the technology as a whole.

Weil, et al. (2013) have shown that the composting process renders several pathogens from feces, namely *Listeria monocytogenes*, *Escherichia coli* O157:H7, and *Salmonella* inert. They produce commercially farmed mushrooms safe for human consumption. However, this effect is due to the heat generated through the composting process, rather than remedial action directly by fungi. Thus, the role of heat in such a technology should also be investigated.

The metrics for measuring the efficacy of the fungi at digesting pathogens and waste in a toilet would also have to be determined. This could include pathogen counts, odor, time requirements, and many other factors. A trial and error process could be used with prototypes, informed by collaborating with experts in waste sanitation and compositing toilets, such as Sanivation. Before completing such a project, user research would also have to be gathered to determine if fungi composting toilets are likely to be acceptable to users. This could be completed by surveys, and working with commercial compost toilet companies who may have information on target user preferences and needs. User studies could also be carried out to determine the design and functions that users need – and implementation strategies designed to find barriers and facilitate adoption.

Another consideration that should be investigate is the risk of introducing an invasive, non-native fungi species to a location. This concern could be addressed by seeking out and using only native species from the location the toilet would be used.

One similar project has been founded to create a toilet using mycelium materials, the 'MYCOommunity Toilet' that turns human waste into compost. It was designed by a team led by Professor Dahmen and Professor Hallam at the University of British Columbia, Canada. The design employs a tank made from mycelium materials to contain the solid waste, and divert the urine, designed to be disposed by burying, with the mycelium's purpose being to facilitate the breakdown of human waste. The designers' theorized the tank would fill in 30 days, and then be buried, fertilizing the area buried (Griffin, 2018). The toilet was designed to be a low-impact toilet for refugee camps there was no reported tests on the effectiveness of the toilet to produce fertilizer or verify the epidemiological and environmental impact of the toilet, it is considered an application in its conceptual application. Despite being a positive example of using fungi as a sanitation technology, the toilet does not provide useful materials, beyond being a 'use and bury for fertilizer' solution to sanitation, rather than innovating a new method to deal with waste entirely. It also does not address the issue that the mycelium materials that form the physical structure of the toilet are unsealed, and thus likely to become unsanitary – sealing them would resolve this, but would then slow or prevent the composting action of the container the concept relies upon.

Section Three Summary: Promising Current & Developing Mycelium Materials

In summary, fungi technologies are currently available as packaging, and are being developed as leather replacement technologies. Fungi, particularly coprophilous species, have good potential to be developed into sanitation applications, such as compositing toilets and as a remediation tool. It is also noted that while E Coli was considered as an example of a pathogen that fungi can remediate, human waste contains and attracts many other pathogens (including other fungi). These have yet to be explored or fungi found to address them, and this would be a key part of future research.

SECTION FOUR: CONCLUSIONS & FUTURE WORK

Mycelium materials, made from fungi that have digested waste such as sawdust, are a growing sustainable material option. They provide mechanical properties similar to polymer foams, insulation similar to natural insulator materials such as balsa wood and natural cork, due to having an open-celled structure that resembled the random-fiber mats of synthetic materials. With the addition of natural or synthetic laminate skins, the mechanical properties of the materials can be improved, creating stronger, stiffer materials suited to higher-performance applications. While mycelium materials cannot achieve the mechanical and manufacturing performance of a synthetic polymer, they do offer less-impactful environmental properties as expressed in the Ashby-style slot below in Figure 52:

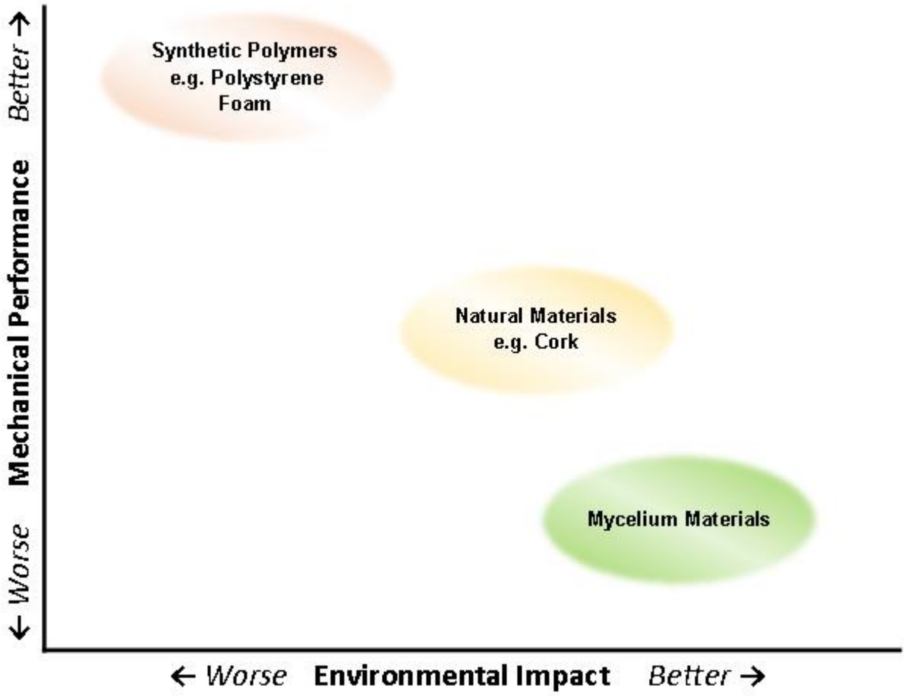

Figure 52. Comparative mechanical and environmental properties of mycelium, natural, and polymeric materials

Mechanical testing according to ASTM testing standards determined the tensile properties, finding an ultimate tensile strength of 176 kPa, with a tensile modulus of 1.3 MPa, and an ultimate compressive strength of 490 kPa. The values for the thermal resistance
thermal conductivity ranged from 0.0014 to 0.0019 m^2 K/W and 0.053 to 0.077 W/m K respectively, which is comparable to current natural insulator materials such as balsa wood and natural cork, and the average maximum use temperature was found to be 390 ºC (734ºF). These properties are comparable to expanded polystyrene foam, suiting the material to non-structural compressive applications, such as packaging and insulation.

Combining the mycelium materials as a core for sandwich composites with carbon fiber and bamboo laminate skins improved the performance from core shear ultimate stress of 36.2 kPa, to 63.3 kPa with bamboo skins and 76.6 kPa with carbon fiber skins, suggesting with development these materials could be suited to higher-performance applications such as interior wall panels and construction bricks.

Testing established that mycelium materials can be shaped using traditional mechanical cutting techniques such as milling and sanding. Finer features and surface decorations can be formed using laser cutting.

Mycelium materials are currently commercialized or being developed for applications ranging from packaging to leather replacements. These materials also show promise for development into sanitation and remediation technologies for use in low-resource settings. Their ability to grow in virtually any environment, and to digest waste to form useful materials, makes them an attractive option to create composting toilets that produce useful by-products.

The hypothesis considered was that mycelium materials can demonstrate the mechanical, thermal, manufacturing, fabrication properties and viable applications, needed for them to be a suitable replacement for synthetic materials such as for polymer foams. Through this work, mycelium materials have demonstrated properties similar to natural materials and synthetic materials. They demonstrated the potential to replace synthetic materials in a variety of non-structural application, such as expanded polystyrene foam for packaging.

Further work is needed to develop these materials and their properties, manufacturing processes and suitable applications. Ideally the information and learning points from the variety of current commercial developments should be shared to help develop the field. Firstly, more types of mycelium materials need to be tested to determine their physical, mechanical, and thermal properties. Different applications require different types of mycelium materials, and researchers and designers could make more informed decision by having more information on mycelium materials properties. Different densities of mycelium materials can achieve different ultimate tensile strengths, stiffness, and compressive ultimate strengths. This should be investigated using tensile, compressive, flexural and other mechanical tests based on ASTM methodologies. As combining the mycelium materials with laminates to form sandwich composites which improved the mechanical properties, further investigation should be carried out to determine the properties when they combined other natural and synthetic laminate skins, such as glass fiber. This would also help inform the material's suitability for various applications that need higher strength materials, where sandwich composites could provide the required performance. Secondly, observations during the growth of mycelium materials highlighted that the geometry of the mold and density of mycelium growth within the mold affects the resulting material density and properties. More research is needed to identify how the mold shape, growing time, feedstock type and many other factors involved in fungi growth, determine the resulting material properties.

In addition, the forming and cutting processes required to shape mycelium materials should also be further investigated. Designers need information on how the material will perform during manufacturing, and while some common processes were explored, investigating more processes would provide further information. Determining the surface finish of the material is particularly important for designers when choosing a material and appropriate manufacturing process, so further investigation of the impact of different spindle speeds, cutting head angles, and other process parameters would also provide more information. Thirdly, while many applications of mycelium materials were briefly mentioned, and three explored using case studies, a deeper exploration into the current and developing applications worldwide is needed. This would help inform researchers and developers about new types of mycelium materials being created. Additionally, to develop sanitation applications that use fungi, such as a compositing toilet, more work is needed to identify which fungi species are suitable to digest human waste. The design of the toilet also needs to be developed, and user research carried out to determine user needs in low-resource environments and help inform the toilet design. Lastly, a key goal of this research effort was to provide researchers and commercial developers with information on the properties, performance and suitable applications of these materials. While this has been achieved, much of the information needed to further this goal is not openly accessible, due to information on the development of mycelium materials often being trade secrets, or intellectual property. Fostering research into mycelium materials, and creating a research community to share research would help develop this emerging research field.

Far-future applications for mycelium materials could include padding materials, growing media, and non-terrestrial applications. As there are millions of species of fungi (Blackwell, 2011), some of which can digest plastics (Maeda et al., 2005), and among many other applications for fungi including for health and nutrition (Stamets, 2005), materials grown from fungi show great potential for use as a waste solution, and also for in-situ remediation of soils contaminated with heavy metals (Khan et al., 2000). As fungi materials grow from microscopic spores and digest waste to form extremely large structure, the largest being an *Armillaria ostoyae* species the size of four square miles in eastern Oregon (Casselman, 2007), they are suited to future applications where there is very limited space to transport the material seeds, but continuous supplies of feedstock. This lends them future development a materials for use in space, as thousands of species each with its own specific use, could be transported into an extraterrestrial environment, and grow from the waste there. Finally, as consumers become increasingly conscious of the environmental and health impacts of materials, materials which can provide health benefits and reduced environmental impact are likely to continue to be developed, creating opportunities for mycelium materials.

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