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Review

Physico-Mechanical and Thermodynamic Properties of Mycelium-Based Biocomposites: A Review

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Abstract: Reducing the use of non-renewable resources is a key strategy of a circular economy. Mycelium-based foams and sandwich composites are an emerging category of biocomposites relying on the valorization of lignocellulosic wastes and the natural growth of the living fungal organism. While growing, the fungus cements the substrate, which is partially replaced by the tenacious biomass of the fungus itself. The final product can be shaped to produce insulating panels, packaging materials, bricks or new-design objects. Only a few pioneer companies in the world retain a significant know-how, as well as the ability to provide the material characterization. Moreover, several technical details are not revealed due to industrial secrecy. According to the available literature, mycelium-based biocomposites show low density and good insulation properties, both related to acoustic and thermal aspects. Mechanical properties are apparently inferior in comparison to expanded polystyrene (EPS), which is the major synthetic competitor. Nevertheless, mycelium-based composites can display an enormous variability on the basis of: fungal species and strain; substrate composition and structure; and incubation conditions. The aim of the present review is to summarize technical aspects and properties of mycelium-based biocomposites focusing on both actual applications and future perspectives.

Keywords: mycelium; fungi; biocomposite; foam; sandwich; lignocellulose; physical properties; mechanical properties; thermal properties

1. Introduction

Due to the increasing demand for "green" materials and productive processes, extensive literature has been developed about the so-called biocomposite and bio-based materials. Such terms basically aim to highlight the derivation of the raw materials from biological sources, although the intervention of a further biological activity by living organisms is not excluded in the transformation process of the materials themselves. Far from being a univocal definition, biocomposites have been referred to by Dicker et al. (2014) [1] as composite materials, where a bio-polymer or bio-derived polymer is reinforced by natural fibers. It should be noted that natural fibers are completely or partially constituted of biopolymers. Moreover, the term "fiber" has a different meaning in chemistry, materials engineering and botany [2–5]. Analogously, bio-based materials are defined as materials containing at least one component that is biologically produced and completely biodegradable [6]; thus, such a definition does not exclude the presence of other synthetic components [7].

A major attractive factor of biocomposites is the possibility to exploit biological wastes and/or residue, such as husks, waste fibers, and residual stems. Waste and residues are thus valorized rather than discarded, according to the principles of the circular economy [6,8].

Early explorations in the use of fungi as biomaterials began during the 1990s by the Japanese scientist Shigeru Yamanaka, who researched mycelium for the production of paper and building materials [9]. Since then, mycelium-based composites have been investigated for commercialization [10,11] and explored for their potential in several conceptual projects (such as the recent exhibition "Fungal Futures": www.fungal-futures.com, 2016, NL) [12]. Research in Europe on mycelium-based composites has been initiated by Maurizio Montalti, in collaboration with Utrecht University (the Netherlands), testing mycelium technology with the local waste streams of the European agricultural industry. This experimentation was funded first by the Creative Industry Fund NL (Stimuleringsfonds) and later from national and European institutions for research (NWO, EEC-H2020) [13,14].

Fungi (apart from yeasts) are organisms able to give cohesion to incoherent materials due to the production of a mass of microscopic filaments (called hyphae) that form the mycelium. Fungi never produce true tissues; nevertheless, hyphae can display different types and specializations related to substrate degradation and the development of reproductive structures [15,16]. According to the available literature, mycelium in biocomposites is coupled almost exclusively with other (non-fossil) materials derived from biological processes, such as plant materials, to exploit the natural growth of the fungal organism on these substrates. Since the variability of the degradative processes in fungi is enormous, lignocellulosic substrates are colonized by a plethora of species. Nevertheless, the most efficient degradation of the lignocellulosic compound is well-known to be carried out by wood-decay fungi (WDF) mostly belonging to Basidiomycota. Wood-decay species can degrade cellulose, hemicelluloses and lignin by enzymatic or non-enzymatic mechanisms, of which selectivity is both species-specific and environmentally determined [17]. The substrate matrix thus is forced and penetrated by the hyphae, which develops inside as an increasingly tight net. Over time, the substrate is replaced partially by the fungal biomass and the resulting mycelium is able to strongly cement the substrate itself, resulting in a biocomposite material (Figure 1).

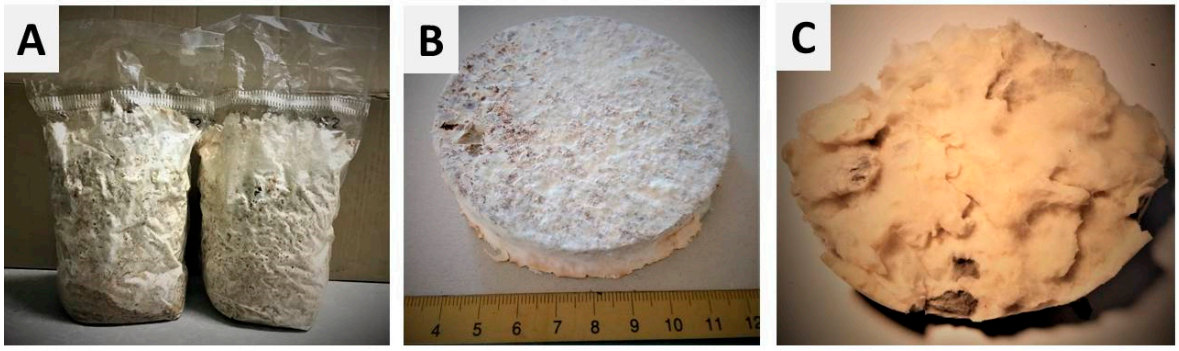

Figure 1. Cultivation bags with mycelial growth in progress (**A**) and final biocomposite (**B**,**C**). Pictures by C. Girometta and R.M. Baiguera.

The fungal colonization of the substrate is often inhomogeneous as confirmed by SEM imaging (Figure 2).

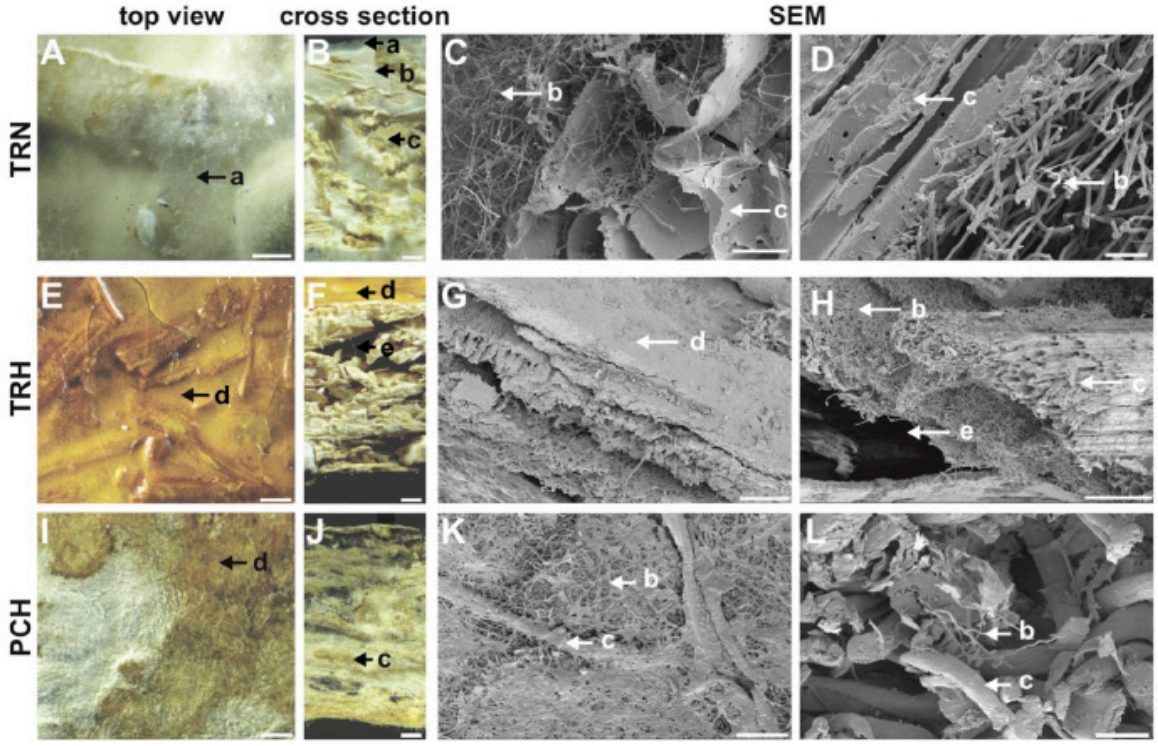

Figure 2. Stereomicroscopy and cryo-SEM images of different biocomposites. TRN (**A**–**D**) = *T. multicolor* on straw without heat pressing; TRH (**E**–**H**) = *T. multicolor* on straw with heat pressing; PCH (**I**–**L**) = *P. ostreatus* on cotton with heat pressing. Arrows indicate aerial hyphae (a), mycelium (b), substrate (c), fused hyphae (d) and air-voids (e). Scale bars represent 1 mm (**A**,**B**,**E**,**F**,**I**,**J**) 100 µm (**H**), 50 µm (**C**,**G**,**K**), 20 µm (**L**) and 10 µm (**D**). Reprinted with permission of Apples et al. (2019) [18].

The surprising performances of mycelium-based biocomposites have been explored particularly to produce new materials for packaging, thermal and acoustic insulation and a broad variety of design objects and furniture [19–21].

Biocomposites production involves low embodied energy, the resulting materials are biodegradable, and they have a potential for cost-effectiveness [22,23]. Few recent research studies have demonstrated the competitive performances of non-pressed, foam-like mycelium-based composites, compared to conventional materials like expanded polystyrene (EPS) or other foams [6,24,25] or to other bio-based composites such as hempcrete [6]. Altogether, mycelium-based materials have demonstrated their potential to replace the use of less environmentally friendly materials, such as bioplastics or wood composites [26]. The resulting materials are fully natural and compostable, thus potentially supporting the transition toward a circular economy, where the value of materials and resources is maintained in the economy for as long as possible and the generation of waste is minimized (EU action plan: COM/2015/0614) [27]. Moreover, a unique feature of mycelium-based composites is the wide diversity of technical and aesthetic properties that can be achieved through minimal variations in the fabrication process, such as the substrate or fungal strains employed. Only a few companies in the world have gained a truly competitive know-how and the economical/technical faculty to provide the complete characterization of the products however. Moreover, several details concerning the material technology and the productive process are kept covered by industrial secrecy and are not published in any paper. Thus, technical literature provided by reviewed publications is quite poor, whereas greater attention has been paid to design and ethical aspects.

The aim of the present review is to critically summarize the available knowledge pertaining to the physical, mechanical and thermodynamic characterization of mycelium-based biocomposites with regard to the actual and potential applications. This work can be useful to direct future research

activities to obtain new materials that can be used for increasingly diversified applications, particularly in packaging, building insulation and interior design.

2. Materials and Properties

According to the species, fungi can degrade the cellulose or lignin compound of plant biomass in a preferential manner, whereas hemicellulose is usually attacked by all the species. Most fungi are selective and degrade both cellulose and lignin, despite being shifted toward a cellulose preference or lignin preference. Such a preference is both species-specific and environmentally determined [17,28,29].

The variety of methods in the productive process of fungal biocomposites basically relies on the colonization of a substrate that is shaped contemporarily or subsequently to the mycelial growth. Once the colonization is carried out, the biocomposite material is pressed and dried by different protocols of pressure and temperature, as shown in Figure 3 [30].

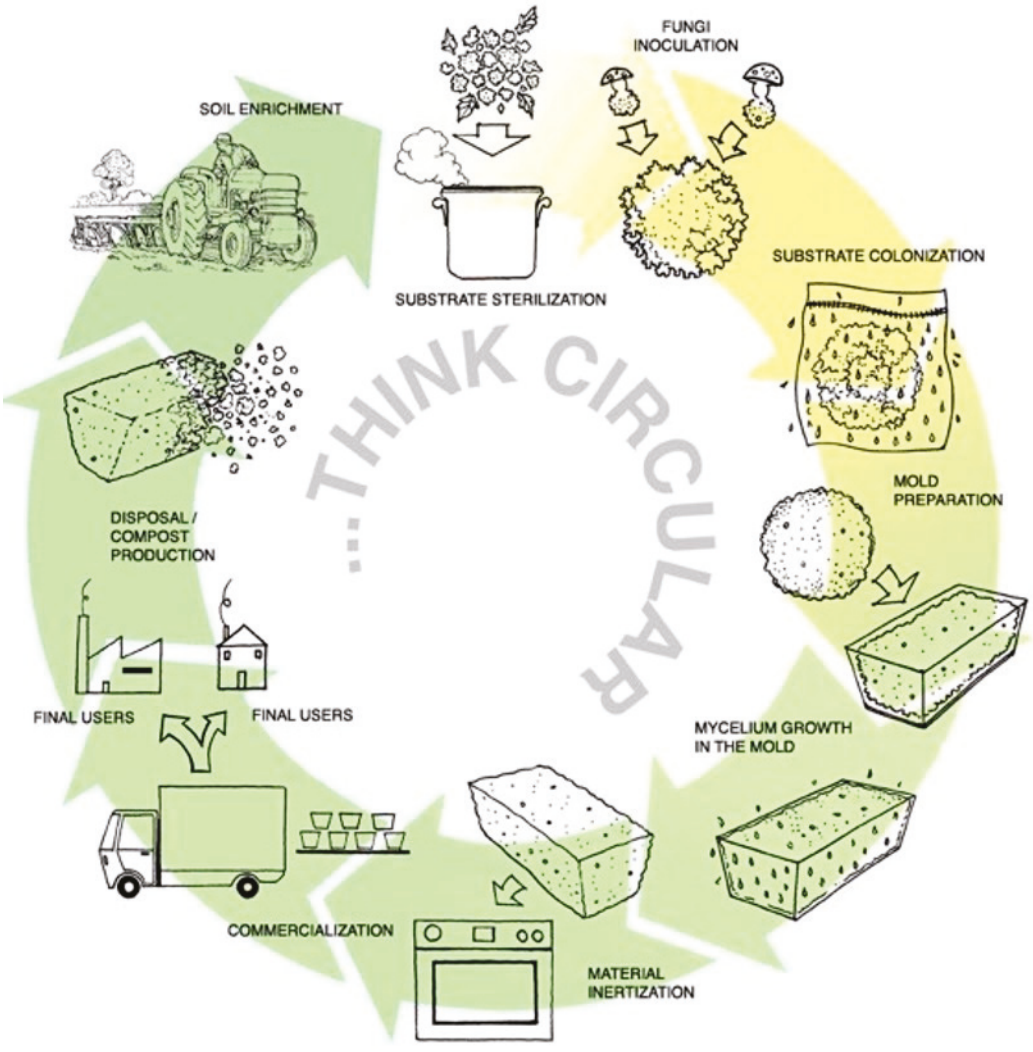

Figure 3. Diagram showing the production and life cycle of mycelium-based materials. The picture also shows how these materials comply with circular thinking. Reprinted with permission of ©MOGU, https://www.mogu.bio/ [31].

A strict categorization is not possible, however, as only two main categories of mycelium-based composites have been developed: foams (Figure 4) and sandwich composites (Figure 5). Mycelium-based foams (MBFs hereafter) are low-density materials emerging from the colonization of

a lignocellulosic substrate by the fungus. This results in a highly anisotropic, fibrous structure where the fungal biomass and residual substrate coexist in a unique matrix, as previously discussed.

Discussed in material science, "solid foam" is usually the result of gas bubble dispersion into a solid matrix. Mycelium-based composites do not fit exactly into such a definition but, despite this, they are often placed in the foam category due to their high porosity and slight rigidity.

Basically, MBFs lack final pressing (either heat or cold); nevertheless, the biomaterials referred to as "mycelium bricks" or "panels" can be included in the category of MBF, although pressing and density can be notably higher with respect to other products [32–35].

Figure 4. Typical structure of mycelium-based foam. Reprinted with permission of Karana et al. (2018) [36].

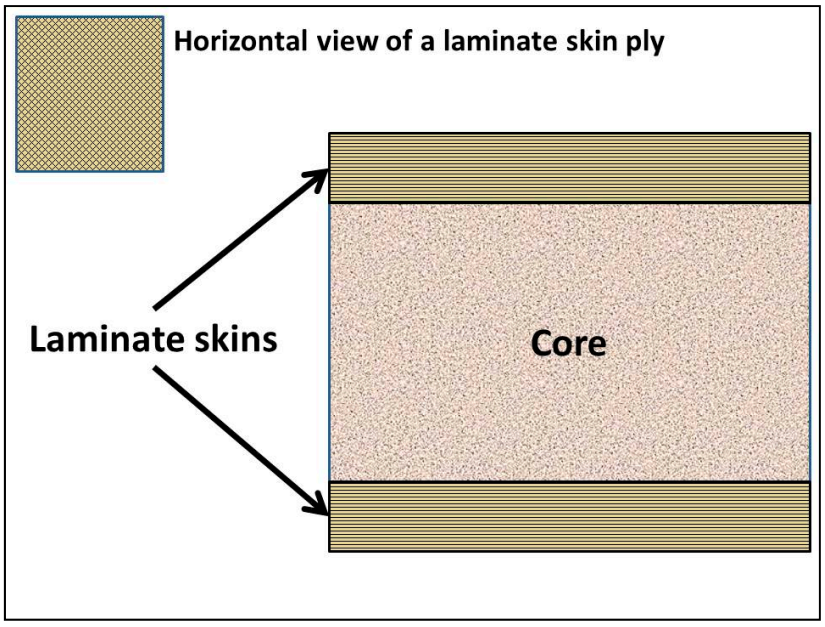

Figure 5. Typical structure scheme of a mycelium-based sandwich composite. Modified from Jiang et al. (2017) [22].

Sandwich composites are characterized by a scaffolded structure where the external layers are jointed due to the gluing action of the mycelium growing in the core material. Additional glue can be added to enhance the material cohesion. The core material is similar to an MBF included between two layers of lignocellulosic material, which are colonized in turn and glued to the core itself, resulting in a single block.

Figure 5 shows laminate skins are made of a variable number of plies, to condition for colonization by mycelia and glue perfusion (if applied) to be unhampered [37,38].

The numeric values reported for physical, mechanical and thermal parameters in both MBFs and mycelium-based sandwich composites (MBSCs) are reported in Table 1.

Table 1. Physical, mechanical and thermal values available in literature for mycelium-based composites. MBF = mycelium-based foam; MBSC = mycelium-based sandwich composite.

Density (g cm^{-3})	Thermal Conductivity (W $m^{-1}K^{-1}$)	Young's Modulus (MPa)	Compressive Strength (kPa)	Flexure Strength (kPa)	Tensile Strength (kPa)	Material	Ref.
0.183 ± 15.1	-	-	41.72 ± 13.49	10.91 ± 4.41	49.90 ± 20.00	MBF	[24]
0.25	-	-	-	-	-	MBF	[34]
0.05–0.06	0.078–0.081	-	-	-	-	MBF	[35]
0.10–0.14	-	66.14–71.77	670–1180	-	100–200	MBSC	[37]
0.16–0.28	-	-	-	-	-	MBF	[39]
0.07–0.22	0.10–0.18	123–675 *	1–72 *	7–26 *	-	MBF	[40]
0.10–0.24	-	2–97	-	50–860	10–240	MBF	[18]
0.16–0.28	0.05–0.07	5.39–58.63	29–567	-	-	MBF	[41]
-	4.27–8.35	-	-	-	-	Other	[42]
0.3–0.55	-	-	-	-	-	MBF	[43]
0.19–0.59	-	-	-	-	-	MBF	[44]
0.29–0.35	-	-	156–340	-	-	MBF	[45]
0.29–0.34	-	-	125–311	-	-	MBSC	[45]

* Values are not comparable with literature as normalized by the authors to a standard polystyrene density.

2.1. Properties of Mycelium-Based Foams (MBFs)

MBFs have been suggested as competitors for traditional polystyrene and polyurethane foams since they offer a sustainable alternative for small packaging. Despite the production still being limited to a few companies in the world, the productive process is improving quickly, resulting in high-quality materials at reasonably low costs [46,47].

2.1.1. Density

The low density is regarded as a major factor in the competitiveness of MBFs (Table 1). Besides the desirability of low weight for packaging purposes, density affects other important material properties, both from a physical-mechanical and a thermodynamic point of view.

The control of density and its homogeneity within the material are still problematic aspects in MBFs, as it is strongly related to substrate composition and structure. Logically, the higher the fraction of grains in the substrate, the higher the final density is in comparison to protocols that increase the fraction of fibers, husks or wood pulp [39,48]. Correspondingly, Holt et al. (2012) [40] observed that density can be lowered by using liquid cultures for inoculum (rather than grain spawn) and by avoiding particles over 2 mm in diameter in the substrate.

Since the density of natural fibers ranges between 1.2 and 1.5 g cm^{-3}, and the density of wood is between 0.3 and 0.88 g cm^{-3}, the decrease in density of the final MBF strongly depends on the porosity [43]. It is interesting to note that Islam et al. (2017) [49] reported a density of 0.03–0.05 g cm^{-3} for a novel biomaterial made of compressed mycelium only. This value is quite similar to EPS which has a density of 0.022–0.030 g cm^{-3}. This means that the replacement of plant biomass by fungal biomass decreases the density of the composite material.

Apart from fungal colonization and biomass replacement, pressing (either cold or heated) is a major variable affecting the final density in MBFs. Panels typically are shaped by pressure of about 30 kN, which obviously increases the density itself in comparison to non-pressed materials [18]. A significant proficiency concerning mycelium-based materials is retained by a few companies and laboratories only, which represents the best available technology (BAT). Based on the literature reviewed here, BAT results in a density for pressed MBFs that is approximately 20% of non-EPS and 17% of commercial natural fibers, as shown in Table 1.

2.1.2. Thermal and Acoustic Insulation

Foams are generally regarded as good insulators and MBFs also have been suggested for thermal insulation, such as to produce panels to place in a wall core (Figure 6), and they are the most cited parameter in literature [33].

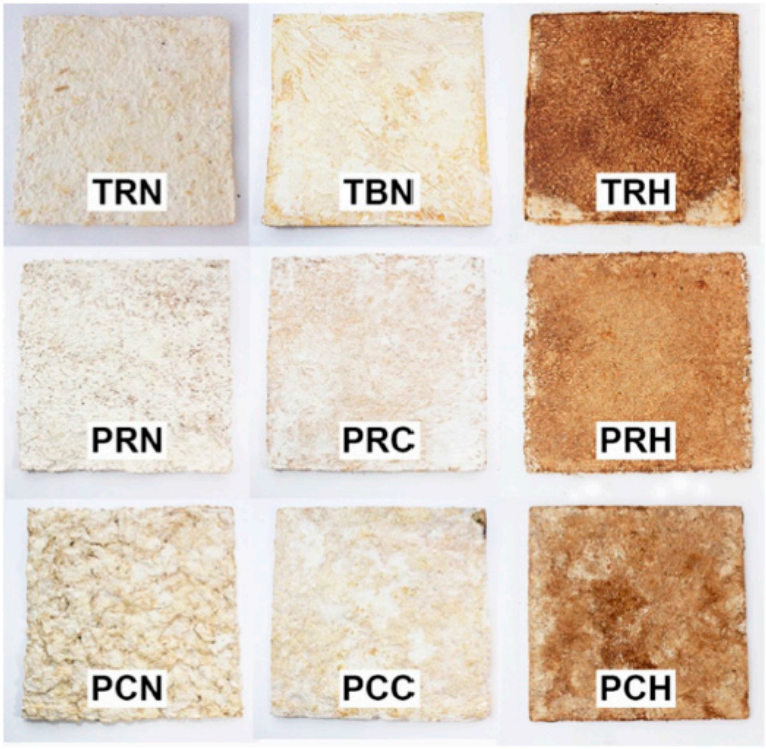

Figure 6. Panels from growth of *Trametes multicolor* and *Pleurotus ostreatus* on different substrates. TBN = *T. multicolor* on sawdust; TRH = *T. multicolor* on straw with heat pressing; TRN = *T. multicolor* on straw without heat pressing; PCH = *P. ostreatus* on cotton with heat pressing; PCC = *P. ostreatus* on cotton with cold pressing; PCN = *P. ostreatus* without pressing; PRH = *P. ostreatus* on straw with heat pressing; PRC = *P. ostreatus* on straw with cold pressing; PRN = *P. ostreatus* on straw without pressing (PRN). Reprinted with permission of Apples et al. (2019) [18].

Aside from the thermal conductivity, the highly specific heat capacity (7.4–10.2 kJ kg^{-1} k^{-1}) also has been reported in comparison to commercial materials [35,50–53].

The development and characterization of a novel, fungus-based material for thermal insulation resulted in a patent in the USA by Amstivslavski et al. (2017) [41] on the basis of the results previously published by Yang et al. (2017) [48]. The authors developed a scaffolded structure by reiterated layer deposition, each layer being constituted by a colonized substrate. Inter-layer cohesion is provided by the cementing action of the mycelium growth. Table 1 shows the thermal conductivity is the lowest discussed in literature, particularly in the dried samples. While a residual moisture fraction usually is tolerated for other applications of MBFs, this work notes the need for complete drying to improve the

insulation properties. Curiously, Holt et al. (2012) [40] reported higher conductivity values, despite the density of the tested material being lower. Velasco et al. (2014) [42] correspondingly reported the evidence of a decrease in the thermal conductivity of clay bricks when adding spent mushroom compost (SMC) over 10%. Nevertheless, the data provided by the same authors about the SMC itself are unexpected and quite confused, lacking a unity of measurement, moisture percent in the tested sample and directly verifiable sources.

MBFs, as a whole, have good potential as thermal insulators in comparison to a wide variety of commercial materials commonly adopted in engineering, yet the thermal conductivity is generally higher. MBFs, by relying on the BAT, already can be regarded as competitive towards unconventional insulation materials, although a decrease of about 33% in thermal conductivity is required to be fully competitive with commercial materials [50,54].

Besides thermal insulation, MBFs have been tested for acoustic insulation as well. Pelletier et al. (2013) [43] tested panels using different substrates and reported an acoustic absorption of over 70–75% at 1000 Hz, even by the worst performing samples. Comparisons among audio spectra show the highest absorption occurred when the substrate was composed of 50–50% switchgrass–sorghum. Since the study focused on damping the dominant road frequencies, intra-wall panels might be applied for joint thermal-acoustic insulation [46]. Extra wall insulation has been proposed as well, as the surface roughness in panels increases acoustic absorption (Figure 7).

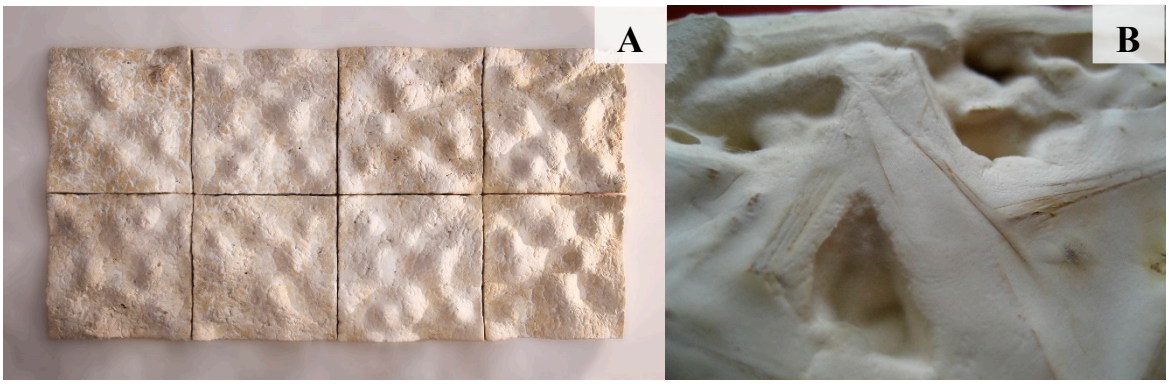

Figure 7. Comprehensive look (**A**) and detail (**B**) of Mogu 3D wave panels for acoustic insulation. Reprinted with permission of ©MOGU, https://www.mogu.bio/ [31].

2.1.3. Thermal Properties and Fire Safety

Only a few studies deal with the behavior of the final mycelium-based composites toward fire, combustion and/or pyrolysis. According to Jones et al. (2018) [44], MBFs show time that is similar to ignition in comparison to extruded polystyrene (XPS) foam, but significantly shorter than particleboard. Ignition temperature has been reported to range between 200 °C and 400 °C but, interestingly, mycelium does not show intrinsic flame-retardant properties and only acts as a cement instead [22,49]. According to Jones et al. (2018) [44], this is due to the passivation of the MBF occurring when its superficial layers turn to char (primarily amorphous carbon). Char is, in fact, known to delay smoke production and spread, as well as decreasing thermal conductivity [55].

Moreover, the composition itself of the fungal cell wall suggests a potential improvement in fire properties in mycelium-based biocomposites with respect to plant materials only.

The chitin component of the fungal cell wall probably is regarded as a pivotal factor. Chitin is a polymer of N-acetyl glucosamine, of which long chains aggregate in turn to make fibrils. Chitin fibrils have been reported to build the main basal layer of the cell wall, yet they are also bonded to all the macromolecules in the upper layers. Aside from H-bonds, chitin has been reported to be covalently bonded to polysaccharides and thus to peptides [56,57]. Chitosan represents the de-acetylated form of chitin; yet, acetylation and de-acetylation in these polymers are indeed flexible concepts, that

is, 100% acetylation is not expected in fibrils regarded as chitin [58,59]. All told, chitosan seems to be more versatile than chitin for industrial and bioengineering applications; basically, it represents the most important chitin derivative, much easier to obtain and to couple with other molecules as amorphous [60,61]. Several studies report chitosan as a promising additive in flame retardants, mostly relying on the stabilization of materials at high temperatures [62,63]. To cite some examples close to mycelium-based biomaterials, it is known for at least a decade that adding chitosan in the phosphorylation process of cotton fabrics enhances flame retardancy [64]. Analogous results have been reported by Pan et al. (2015), Fang et al. (2015) and Liu et al. (2018) [65–67] with respect to a multi-layer coating made of various chitin derivatives. Layer-by-layer deposition of flame retardants in a chitosan matrix also has been suggested for foams such as polyurethane [68]. According to thermal and thermogravimetric analyses, chitin and its derivatives are more than a matrix embedding flame retardant particles or platelets. Depending on the molecular weight, the main derivative peak for weight loss in chitosan occurs at about 300 °C, whereas chitin peak is submitted by most authors to occur at about 350–400 °C [69–72]. To compare, the polymers building the main fraction of the plant substrates in mycelium-based biocomposites have a lower thermal stability. To be exact, pure crystalline cellulose has a narrow main peak at about 350 °C, which is higher than hemicelluloses; the latter, moreover, display a broad range for weight loss (200–375 °C) [73]. Thermal degradation in lignin occurs through a broad temperature range as its complex molecular structure requires several subsequent steps; however, the main weight loss is usually found in the range of 300–450 °C [74–76]. Moreover, the performance of an intimate mixture of the mentioned molecules, such as a typical plant stem, behaves differently with respect to its separate components; that is, the typical thermogravimetric analysis (TGA) curve for a wood sample shows the major weight loss peak placed at about 500 °C [76,77]. Thus, the cementing action by chitin is expected to positively affect the thermal stability of the mycelium-based biocomposite.

Analogously to chitin, hydrophobins have been suggested as natural flame retardants for fabric coating. Hydrophobins are low-weight proteins found in fungi only; due to their hydrophobic nature, they basically act by self-assembling to create an amphipathic membrane. Found in the fungal organism, hydrophobins are involved in several functions related to cell wall morphogenesis, hydrophobicity and substrate adhesion, both in aqueous and aerial environments [78,79]. Thus, although the comprehensive biomass provided by hydrophobins is negligible, these proteins represent an important driver in the physical properties of mycelia. Correspondingly, it has been reported that *Schizophyllum commune* develops thicker and closer mats if the hydrophobin gene is deleted [80].

Hydrophobins have been reported to favor the production of thermally stable carbonaceous structures (char) when applied on cotton fabrics. The proposed mechanism basically relies on favoring the dehydration of cellulose rather than depolymerization. Reducing the release of volatile species hampers complete combustion and favors charring instead [81].

The heat release rate (HRR) is regarded as particularly important in modelling the growth and spread of fires, whereas TGA can provide additional information on the volatilization of combustion or pyrolysis compounds. While imposing the same incident thermal heat flux (50 kW m^{-2}), Holt et al. (2012) [40] and Jones et al. (2018) [44] reported similar values, both for average HRR (55–75 and 33–107 kW m^{-2} respectively) and peak HRR (66–116 and 79–185 kW m^{-2}, respectively). This means that the peak occurs within the first minute after ignition. Predictably, Jones et al. (2018) [44] reported the highest HRR values when increasing the grain fraction in the substrate, due to its well-known calorific power (1434 kJ kg^{-1}) in comparison to the SMC (about 580 kJ kg^{-1}) reported by Velasco et al. (2014) [42].

According to the same authors, the inclusion of glass powders (an industrial waste) remarkably lowers the whole HRR profile and increases the time to flashover; conversely, glass powder inclusion increases the CO released and should, therefore, be investigated further before application. Jones et al. (2018) [44] demonstrated the CO release displays a highly complex profile as a function of time and tested matrix, that is, different substrates provide different profiles. Specifically, substrates

that are rich in grains display the CO peak at a low temperature and soon decrease due to further oxidation, as consistently observed by Holt et al. (2012) [40].

Although the CO release profile is the most informative tool to test CO emissions, an adequate proxy is the peak position, that is, the time/temperature where the highest CO emission occurs. Due to the mentioned complexity of the release profile, the average emission datum is expected to be misleading instead.

Analogously, the average mass loss rate reported by Holt et al. (2012) [40] is misleading as TGA curves clearly show that mass loss in plant and fungal biomasses typically is concentrated in few temperature ranges. Such ranges are narrow, according to the type of the tested material, and can be negligible in comparison to the overall temperature range adopted to carry out the TGA, which is typically 900 K [82–84].

Based on the above, the mass loss rate should be regarded as complementary information in the light of TGA and especially differential TGA (DTGA). Nevertheless, TGA usually is carried out in very small crucibles containing a few mm^3, (a mass ranging between 2 and 10 mg). Since the substrate colonization and the fungal growth are highly non-homogenous in the millimeter scale, sampling can introduce a significant error when analyzing the biocomposite; thus, separated TGA for substrate and mycelium has been, up to now, preferred [44,82].

2.1.4. Mechanical Properties

The comparison among data reported in literature for mechanical properties (Table 1) is hampered partially due to different standard methods and adjustments [85,86].

Compressive strength is of major concern to test the potential of MBFs as an alternative packaging material and, to a lesser extent, as a building material.

Velasco et al. (2014) [42] reported a linear decrease in break compression strength of fired clay bricks when increasing the percent of SMC, which is actually an MBF; nevertheless, break compression strength no longer decreased when SMC was approximately over 17%. Analogously, brick plasticity linearly increased when the SMC percent increased, but it settled when SMC was over approximately 25% (the exact percent was not provided nor inferable).

According to literature, compressive strength in MBFs consistently ranges between 29 kPa and 567 kPa, the widest range being reported by Amstivslaski et al. (2017) [41]. López–Nava et al. (2016) [24] reported that compressive strength of MBFs is always lower than several standard categories of EPS, except for EPS type XI (ASTM C 578-04).

Nevertheless, Amstivslaski et al. (2017) [41] reported values that are fully competitive with several EPS categories (II, IV, IX, and X) by inoculating *Irpex lacteus* into a more complex substrate, including macerated sawdust pulp, millet grain, wheat bran and a natural fiber. Easily observable, both in nature and laboratory conditions, *I. lacteus* is much more tenacious than *Pleurotus* spp., but less than *Ganoderma* spp.

Despite relying on the results by Yang et al. (2017) [48], reference values in the patent by Amstivslaski et al. (2017) [41] are slightly different, especially for the samples with prolonged incubation.

Holt et al. (2012) [40] conducted a complete physical characterization, including compressive strength. Unfortunately, a comparison with literature is impossible as data by Holt et al. (2012) [40] were normalized to "a standard density of 32.04 kg m^{-3} since this is the density of the polystyrene packaging the EcoCradle material can replace in the market". Since the original density of each sample is not provided, and the normalization procedure itself is unclear, the reader cannot recover a reliable comparison term.

Variability in compressive performance by MBFs apparently is due to both the substrate matrix and the fungal species inoculated. Holt et al. (2012) [40] mostly used milled cotton carpels and cotton seed hulls; the strong cementation of such a material was not banal; nevertheless, mycelium of *Ganoderma* spp. is well-known to display a tenacious consistency. Conversely, relatively low values

obtained by López–Nava et al. (2016) [24] are due to the mycelial features of *Pleurotus* spp., which is significantly softer. Secondarily, the same authors only introduced wheat straw as a substrate, which is not the favorite to maximize the matrix cohesion by fungal growth.

Residual water is a factor in affecting compressive strength and can be due to both absorption by substrate and mycelium. Water absorption reported by López–Nava et al. (2016) [24] at different times is remarkably higher than that characterized by Holt et al. (2012) [40]. This is consistent with the higher water absorption by wheat straw in comparison to cotton carpels and hulls, as well as the highly hydrated aspect of living *Pleurotus* mycelium.

Regarding the mere mycelial component, Haneef et al. (2017) [82] provided an overall investigation on the physical and mechanical properties, just focused on *Pleurotus ostreatus* and *Ganoderma lucidum*. According to the authors, the substrate strongly affects the mycelial composition in polysaccharides, lipids and chitin, as well as the overall morphology and mechanical properties. Mycelium displayed unexpected and apparently contradictory properties in comparison to its composite. Particularly, *P. ostreatus* was found to be stiffer than *G. lucidum* which was suggested to be due to its higher polysaccharidic content.

Islam et al. (2017) [49] tested the properties of a material resulting from the additive overlapping of thin mycelial layers, where all the layers are parallel to support the load. Taking a micro-mechanical point of view, this situation is very different in comparison to the highly anisotropic MBF matrix, as known by the "rule of mixtures" [87]. Alternatively, each mycelial layer itself behaves as an anisotropic matrix, due to the previously discussed structure of the fungal cell wall. Thus, different mechanical behaviors should be expected in a multi-layer mycelial composite and in MBFs. Unfortunately, Islam et al. (2017) [49] do not provide the name of the fungal species under examination, which is covered by industrial secrecy.

Tensile strength of MBFs has been, up to now, poorly investigated, despite the very promising values in comparison to the main standard categories of EPS. The major peculiarity in comparison to other biomaterials is provided by two typical components of the fungal cell wall, more extensively discussed above: chitin and hydrophobins. Pure chitin substrates have been reported to display a tensile strength over 100 MPa by Hassanzadeh et al. (2013) [88]. Furthermore, the major role of chitin in improving tensile performance can be inferred indirectly by considering the results concerning other chitin-based materials, such as "chitin whiskers" and chitin nanofibers. Specifically, 2% chitin whiskers can increase about 200% of the tensile strength in hyaluronan–gelatin nanocomposite scaffolds [89], whereas introducing 10% chitin nanofibers increases the tensile strength approximately 65% in chitosan films [90].

Discussed in the work by López–Nava et al. (2016) [24], MBFs composed of *Pleurotus* and wheat straw reported higher values than all the EPS types. The ultimate tensile strength was reported in the range of 100–300 kPa in the multi-layer mycelial material described by Islam et al. (2017) [49] as it regards to the mycelial component, whereas it ranged between 200 and 1200 kPa according to Haneef et al. (2017) [82]. Moreover, Haneef et al. (2017) [82] reported a smaller elongation at fracture for *P. ostreatus* (4–9%) in comparison to *G. lucidum* (14–33%), since the higher proteic and lipidic fraction in *G. lucidum* would act as a plasticizer. The aforementioned author basically refers to structural proteins occurring in the cell wall; nevertheless, Appels et al. (2018) [80] demonstrated that the deletion of the hydrophobins gene in *S. commune* indirectly enhances the tensile strength, as the fungi respond by producing a closer and denser mycelial mat. Thus, Appels et al. (2018) [80] suggest that the mechanical behavior of mycelium is more similar to thermoplastics when hydrophobins are not produced [91].

According to the same authors, the availability of less recalcitrant substrates (specifically, potato-dextrose rather than cellulose) would stimulate the production of the aforementioned plasticizers rather than rigid polymers. Nevertheless, the substrate was not found to significantly affect the ultimate strength (0.7–1.1 MPa). This is consistent with Appels et al. (2018) [18], who tested tensile strength, both in panels produced with *P. ostreatus* and *Trametes ochracea* (referred by the authors as *T. multicolor*), by different protocols. The authors found that tensile strength was affected by neither the

species nor substrate; they pointed out the pressing instead was the major factor, and specifically, heat pressing was reported to increase tensile strength in comparison to cold pressing or non-pressing.

Flexural strength is the stress at fracture from a bend or flexure test; it is also known as bend strength, modulus of rupture or transverse rupture strength. This parameter is ambiguous since it originally was conceived for brittle ceramics, graphite and semiconductors as a surrogate for tensile strength. Specifically, it should be noted that: a) flexural strength decreases by increasing the specimen size and consequently the crack probability; b) values attributed to flexural strength are higher than corresponding values of tensile strength because only a fraction of the specimen volume is subjected to tensile stress when performing a flexure test; c) porosity strongly contributes to lower flexural strength, mainly because the load is applied on a smaller cross-sectional area [92].

Flexural strength of MBFs was investigated by both López–Nava et al. (2016) [24] and Holt et al. (2012) [40]. The last ones did not provide comparable data. According to López–Nava et al. (2016) [24], values of MBF are significantly lower (6–63 folds) than all the compared EPS categories. Unlikely asserted above (b), the same authors found higher values for tensile strength than for flexural; this is consistent with the remarkable porosity in mycelium-based biocomposites (both MBF and MBSC), as asserted in (c).

Values reported by Appels et al. (2018) [18] are remarkably higher than EPS [91]. Consistently with (b), the same authors refer to flexural strength values higher than for tensile strength; concerning this peculiar (non-ceramic) material, the authors suggest this is due to the remarkable elasticity of fungal skin in comparison to the substrate (Figure 8). Consistently, Tudryn et al. (2018) [93] report a positive correlation between flexural stress at yield (as well as bulk flexural modulus) and branch density. Since different protocols were used in the mentioned study, it is moreover observed that flexural strength increased from non-pressed to cold-pressed and hot-pressed panels. Since pressing decreases the porosity, this is consistent with (c).

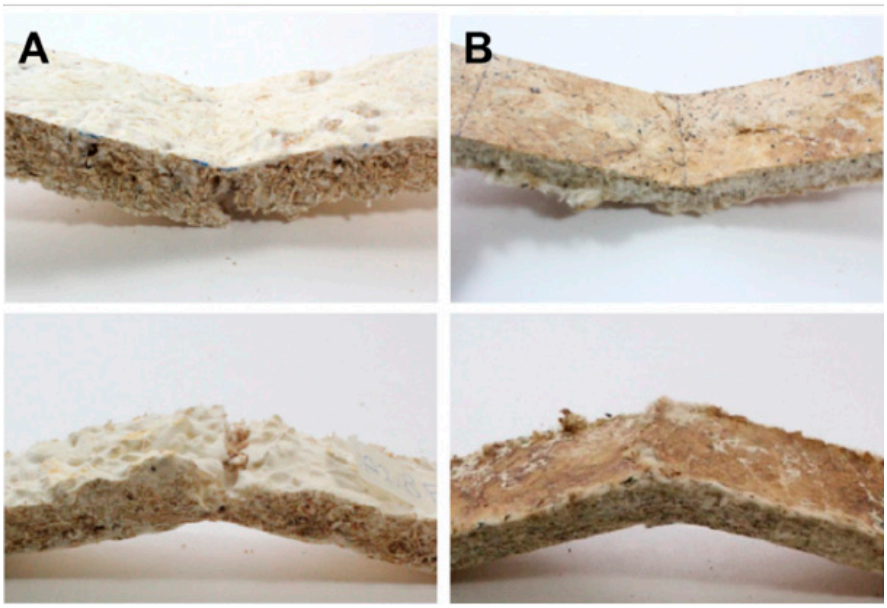

Figure 8. Higher resistance of the fungal skin in comparison to the substrate. The top side remains intact under flexural stress whereas the bottom side is broken. (**A**) *T. multicolor* on straw without heat pressing; (**B**) *P. ostreatus* on cotton with heat pressing. Reprinted with permission of Apples et al. (2019) [18].

The elastic deformation of materials is usually expressed by means of shear modulus and Young's modulus.

The patent by Amstivslaski et al. (2017) [41] relies on the data by Yang et al. (2017) [48]; authors observed shear failure in densely packed MBF samples, whereas bulging failure occurred when the

same stress was imposed on loosely packed samples. Shear moduli observed by the authors were lower than Young's moduli, except for an overlapping range between 5.39 and 19.20 MPa. Both shear modulus and Young's modulus were always significantly higher in MBF than in EPS [94]. Consistently, Haneef et al. (2017) [82] reported Young's modulus in the range of 3–11 MPa and 17–27 MPa for *G. lucidum* and *P. ostreatus* pure mycelia, respectively. The two species are clearly different in the consistency of the mycelial mat, *G. lucidum* being more tenacious; yet, the chemical characterization performed in the same study did not satisfactorily clarify the relation between chemical composition, structure and elasticity. Chitin is a striking factor in affecting the mechanical performance of biocomposites; by testing different dry substrates, elastic modulus in chitin was reported to be 2.5 GPa by Hassanzadeh et al. (2013) [88]. Despite being conducted on basically different materials, several studies provide a glimpse of this concern. The introduction of chitin nanofibers in chitosan films, gelatin methacryloyl, has been reported to significantly increase Young's modulus [88,90] for example. Additionally, it has been suggested that the synergic interaction between chitin and other polymers can increase elasticity with respect to the separate components [95]. An increase in mycelial mat tightness and density has been reported to positively affect the Young's modulus by Appels et al. (2018) [80]. Notably, the same authors observed such an increase in mycelial mat density as a response to deletion of the hydrophobin gene. Hydrophobin-lacking fungal organisms might be enhanced to behave more like thermoplastics, from a mechanical point of view.

The species under examination by Yang et al. (2017) [48], that is, *I. lacteus*, displays more similar features to *G. lucidum* than to *P. ostreatus* [91]; yet, a characterization of *I. lacteus* pure mycelium is not available. Surprisingly, Appels et al. (2018) [80] did not find a correlation between species (nor substrate) and elastic modulus of panels, although the mycelial consistency of the tested species is very different [96]. As mentioned above, concerning tensile strength, they suggest heat pressing as a striking factor in elastic modulus increase.

2.2. Mycelium-Based Sandwich Composites

An MBSC is constituted by a core and two laminate skins, the former resembling an MBF and the latter usually made of multiple layers of fabric or mycelial mat.

A critical discussion on the productive process and cost modeling for a typical manufacturing system dealing with MBSC has been provided by Jiang et al. (2017) [22], and is basically summarized in the following points: (1) mycelial mat or fabric is cut according to the desired shape to produce the plies for laminate skins; (2) plies are impregnated with natural glue compatible with fungal growth; (3) treated plies are shaped, sterilized and solidified; (4) mycelium is allowed to colonize the plies to cement the laminate skins; (5) mycelium is inactivated by drying; (6) if necessary, natural resin is infused to reinforce stiffness; (7) 23.34% of raw materials is discarded with respect to the final product and glue infusion is the most critical step due to the high failure rate.

Concerning the above discussed biofoams, MBSCs present an additional heterogeneity factor as laminate skins and core are very different materials, from a mechanical point of view.

Jiang et al. (2017) [22] show the fabric selected for laminate skins remarkably affects the fungal colonization and consequently the mechanical properties. The authors, in fact, report a more efficient colonization and a higher biomass production when using flax rather than jute or cellulose, at least in the species under examination (unreported as covered by industrial secrecy). Thus, the fungus was able to strongly cement the fabric layers, both to each other and to the core, by developing a tight mycelial net.

Consistently, the ultimate strength and yield stress are almost double in samples made with flax skins than in samples made with jute or cellulose skins. Specifically, ultimate strength has an average value of almost 35 kPa in a flax MBSC, whereas jute and cellulose MBSCs have respective ultimate strength values of 20 and 16 kPa; yield stress has an average value of 27 kPa in a flax MBSC, whereas jute and cellulose MBSCs have respective ultimate strength values of 12 and 15 kPa.

Yield stress and ultimate strength studied in the same work are only referring to the core and represent "the stresses above which permanent deformation and failure occur due to shearing". Yet, the laminated skins are susceptible for interlaminar shear failure, which was reported as the most common failure beside core tensile.

Ziegler et al. (2016) [37] reported an MBSC with a core made of hemp pith and cotton mat, whereas the surface binding fabric is made of a generic "natural fiber fabric such as burlap".

Table 2 displays MBSCs showing considerably higher elasticity modulus, compressive strength and, to a lesser extent, tensile strength in comparison to MBFs. This is due to the double compression of the material (in the core colonization stage and after skin colonization) and to the reinforcement operated by the laminated skins. Consistent with observations by Jiang et al. (2017) [22] concerning failure modes, the increase in tensile strength is related to the presence of the laminated skins, and yet the cementing action by mycelium might provide additional elasticity to the skins themselves. Thus, an efficient colonization of the laminated skins might reduce the interlaminar shear failure. Nevertheless, it should be noted that the substrates used by both Jiang et al. (2017) [22] and Ziegler et al. (2016) [37] have a fibrous structure that favors the fungal colonization and the core cohesion, different from the incoherent material (hulls, straw) often introduced in MBFs. Additionally, the relative recalcitrance of any natural fiber to fungal degradation stimulates the fungus itself to develop a more elastic mycelium, consistent with the previously discussed Haneef et al. (2017) [82]. The strong relation among skin colonization, achievement of fungal biomass and cementation of both core and skin layers is suggested by Ziegler et al. (2016) [37] to be pivotal to explain the compressive properties. Besides elasticity, the low compressive strength reported by the authors was related to the porous structure, insufficiently colonized.

Table 2. Drying protocol and water uptake in dried samples. Semicolon indicates successive steps within the same treatment. MBF = mycelium-based foam; MBSC = mycelium-based sandwich composite.

Drying Protocol		Response to Moisture Exposure		Response to Water Immersion		Material	Ref.
T (°C)	Time (h)	Relative Humidity (%)	Weight Increase (%)	Time (h)	Weight Increase (%)		
150; 80	0.20; 24	60–80	3.15–11.63	192	43–508	MBF	[18]
20; 80	0.20; 24	60–80	3.74–10	192	238–262	MBF	[18]
80	24	60–80	5.80–10.96	192	246–281	MBF	[18]
60	8	-	-	3	10–48	MBF	[40]
60	8	-	-	75	6.4–30.7	MBF	[40]
60	8	-	-	168	93.5–198	MBF	[40]
?; 25	48; 48	-	-	Not reported *	114.1–278.9	MBF	[24]
82; 93; 250	12; 8; 0.20	-	-	-	-	MBSC **	[38]
110	24	-	-	50	298.7	MBSC	[37]
110	24	-	-	>50	340.3–350.5	MBSC	[37]

(*) Authors refer to ASTM D 570-98(1985). (**) Data refer to the core material only.

Altogether, all the compressive, elastic and tensile parameters reported by Ziegler et al. (2016) [37] for MBSCs are remarkably lower than the corresponding wood–polymer composites, balsa or expanded polystyrene. Conversely, ultimate shear strengths reported by Jiang et al. (2017) [22] are comparable with Styrofoam™ (EPS), although the density of the latter is 59–83 folds lower than the mentioned MBSC.

Since a linear relation occurs between density and shear strength in EPS, this material seems to be highly superior than actually available MBSCs, apart from environmental considerations [97,98].

2.3. Moisture as a Critical Factor in Affecting Properties in Mycelium-Based Composites

As mycelium-based composites result from a biological process, an adequate moisture in the substrate is needed to allow the fungus to grow and carry out their physiological functions. Once the substrate colonization is supposed to be complete, most of the moisture has to be eliminated to

devitalize the fungal organism and to provide a functional final product. The available literature is not focused on the optimal residual percentage of moisture either in MBFs or MBSCs; alternatively, this parameter is strongly related to the composition of the substrate as well as the fungal species under examination. Thus, the moisture before devitalization is generally believed to be about 59% [42] or 70–80% [16], but the only available reference for an "acceptable" residual percent in the final material is indicated to be about 10–15% [16].

According to the BAT, and relying on commercialized products, moisture is not eliminated completely from the final product. Highlighted by Amstivslaski et al. (2017) [41], exposure to moisture rapidly decreases the performance of mycelium-based composites. Specifically, the same authors mention the negative effect on tension and compression, although they do not provide further data in support. Indeed, moisture was shown to negatively affect elastic modulus by Yang et al. (2017) [48]. Although Appels et al. (2018) [80] did not explicitly investigate the effect of residual moisture on the mechanical properties, their results clearly show that heat pressing increases tensile strength, flexural strength and elastic modulus. Table 2 reports the heating protocol (concerning the same authors) suggests a much more enhanced drying than that occurring by a cool pressing or non-pressing protocol. Thus, drying is related indirectly to the final properties.

Overall, despite the wide gaps in Table 1, the correlation between the drying protocol and mechanical properties apparently is consistent with the rest of the cited literature. A prolonged heating at high temperature thus is related to a high elastic modulus, flexure strength and tensile strength [24,37,41].

Based on the above, the water uptake of final mycelium-based material is of major concern to test the product quality and durability, plus it obviously affects the weight and is a source of deformation. Table 2 shows water uptake both concerns atmospheric humidity and immersion in water, although the latter is more prevalent in testing. According to Haneef et al. (2017) [82], a low water uptake in mycelial mats is consistent with the hydrophobic nature of some fungal proteins and glycol-proteins, such as hydrophobins. Apart from the mycelial mat itself, weight increase strongly depends on substrate; for example, hemp pith uptakes much more water than a fiber cotton down woven mat [37]. Additionally, different coatings might affect water uptake; according to López–Nava et al. (2016) [24], chitosan seems to provide a remarkably lower water uptake than carrageenan and xanthan, although pure dried chitosan also has been reported to be an efficient adsorber [59].

3. Life Cycle Assessment (LCA) and Biodegradability of Mycelium-Based Composites

Mycelium-based technology relies on a biological process very similar to the one occurring in nature, the growth of fungal organisms on lignocellulosic substrates. According to the BAT, edible and/or medicinal macro-species usually are adopted due to their morphological features and cultivability; notably, they are far from being related to the production of mycotoxins or toxic volatile/soluble metabolites [99,100].

Since these species grow on lignocellulosic substrates, a wide variety of agricultural and agro-industrial wastes can be entered as feedstocks, as well as waste fabrics made of natural fibers. Waste stems, part of stems, sawdust, husks, hulls, and peels, among others, provide suitable substrates, depending on the species-specific preference and supply faculty of companies. Overall, such low-cost materials are rescued and valorized as primary sources for feedstock flux [33,101].

Once the substrate optimal conditions are assessed, mycelial growth proceeds autonomously by progressively degrading the substrate until the final steps (drying, pressing, and varnishing if applied).

Mycelium-based technology has been suggested as a model fitting all the pillars of a circular economy according to the following statements [102]:

(1) waste materials are re-entered in a production process rather than being discarded; the final product is in turn combustible or compostable;
(2) pure materials and energy are consumed to create products (panels, and package cases for example) of which post-life has a quality significantly improved in comparison to non-biodegradable products;
(3) a mycelium-based life cycle provides an optimization of resources and energy consumption in comparison to traditional competitor products.

Based on this, the fulfillment of circular economy requirements is strictly related to the product design, as each step of the production process must be consistent with the implementation of sustainability and optimization goals. Regarding mycelium-based technology specifically, the pioneer company Ecovative Design provides an example of successful system analysis where design is a function of the requirements for LCA [36,103].

The main purposes of LCA are self-correction and self-training in design, production process and management through a positive feedback loop. According to the available documentation on the LCA of Ecovative products, three main critical factors are pointed out: (a) inoculum protocol; (b) material configuration comparing different structural feedstocks to increase the strength/weight ratio and to evaluate local supplies (which might lower transport cost); (c) drying protocol, by far the main source (56%) for carbon footprint [104]. Based on the same data, the blend structure is the second most important factor with concern to human health, ecosystem quality and resource consumption. Unexpectedly, a pasteurization process and mixing seem to have a considerably lower impact as they typically behave as a scale cost.

Accordingly, Jiang et al. (2017) [22] report that drying, forming and cutting are the main wrap and failure items in a production process of mycelium-based sandwich structures, apart from gluing. The same authors also highlight labor as a major factor in cost modeling; conversely, mycelium-based technology requires both specialized researchers and workers, as specific automation has not been achieved yet. Silverman (2018) [45] depicted mycelium-based materials with respect to other materials commonly used for footwear soles; according to his LCA, a moderately embodied energy is required to produce these materials in comparison to many plastics, honeycombs and synthetic foams. Interestingly, the same author reports a competitive (and maybe underestimated) ratio between embodied energy for primary production and compressive strength.

Biodegradability of mycelium-based composites has a pivotal function in LCA. As it is stated already, the main competitor of mycelium composites is the EPS. From the chemical point of view, mycelium-based materials are derivatives of polysaccharides, while the backbone of EPS contains aromatics (petroleum-derived). More in detail, an important component in mycelium is chitin, a polymer of *N*-acetylglucosamine that is linked to glucans. The chemical difference is reflected by reactivity; in fact, glycosidic bonds can be degraded by enzimes, thus giving biodegradability, while EPS results are non-biodegradable.

Mycelium-based materials are fully biodegradable on the condition that neither persistent glue, varnish nor coating was added (particularly in MBSCs), although this treatment is expected to significantly improve physical and mechanical properties [105]. To date, natural glues mostly have been tested to prevent the fungus from failing in its growth as a consequence of xenobiotic stress [22].

Although a mycelial mat can be recalcitrant indeed to microbial degradation, cultivation practice reports of frequent contamination by molds into bags, for example by *Trichoderma* spp. and, to a lesser extent, *Neurospora sitophila*, *Penicillium* spp., *Aspergillus* spp. and *Mucor* spp. Apart from the degradation of the plant component (as well as plant-derived fibers), it can be observed that *Trichoderma* is the main model genus representing mycoparasitic fungi due to its chitinolytic system. Since chitin has been estimated to be one of the most abundant biopolymers in the world, the ecological function of chitinases is enormous and they have evolved into a wide family, where fungal chitinases are only a sub-family [106].

Analogously, bacterial degradation and damage by insects (dipteran, coleopteran) and mites contribute to the natural biodegradation of mycelium-based composites [107,108]. It should be noted that composting is meant to occur in moist conditions, where microbial degradation is favored.

4. Conclusions

Increasing attention has been paid to mycelium-based biocomposites as alternative materials to synthetic packaging and insulation panels, as well as developing a new-concept bio-inspired design. Since this is still a pioneering field, standardization in the productive process is still in progress and only concerns the major companies. Overall, a bio-composite is not expected to show the same reproducibility, in comparison neither to synthetic materials nor to mono-component natural materials, as it regards to mechanical and thermodynamic parameters. Apart from the lignocellulosic substrate, the characteristics of a mycelium-based biocomposite are strongly affected by the fungal species selected and its ongoing growth. Thus, the consistency of the mycelium itself is in turn affected by the composition and structure of the substrate. Mycelium-based biocomposites generally rely on a wood-decay fungal species; well-known medicinal species such as *G. lucidum* and *P. ostreatus* have the advantage to be cultivated and a rich technical literature is, therefore, available to optimize the growth conditions. Other species such as *I. lacteus*, mostly neglected by non-mycologists, have reported very promising results.

Nevertheless, the literature providing a satisfactory material characterization is still scarce. Due to the wide range of available parameters, results are often incomparable among different studies. Poor discussion is often provided about the results of the contingent study itself, as well as the comparison to other conventional materials, such as EPS. This usually is regarded as the main synthetic competitor to mycelium-based biocomposites, but the latter appears inferior as it concerns several physical-mechanical parameters; as these two materials are completely different, more investigation would improve the technical impact of the publications.

To conclude, mycelium-based materials, of which exploration has only recently begun, have great potential. The improvement in know-how is expected to particularly improve the mechanical properties and to standardize the productive process, whereas insulation and thermal properties already have shown competitive results.

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