

Novel Mycelium-Based Biocomposites (MBB) as Building Materials

Zinta Zimele^{1,*}, Ilze Irbe², Juris Grinins², Oskars Bikovens², Anrijs Verovkins² and Diana Bajare¹

 ¹Riga Technical University, Riga, LV-1658, Latvia
 ²Latvian State Institute of Wood Chemistry, Riga, LV-1006, Latvia
 *Corresponding Author: Zinta Zimele. Email: zinta.zimele@edu.rtu.lv Received: 15 January 2020; Accepted: 21 May 2020

Abstract: Novel mycelium-based biocomposites (MBB) were obtained from local agricultural (hemp shives) and forestry (wood chips) by-products which were bounded together with natural growth of fungal mycelium. As a result, hemp mycocomposites (HMC) and wood mycocomposites (WMC) were manufactured. Mechanical, water absorption and biodegradation properties of MBB were investigated. MBB were characterized also by ash content and elemental composition. The results of MBB were compared with the reference materials such as the commercial MBB material manufactured by Ecovative® Design (EV), hemp magnesium oxychloride concrete (HC) and cemented wood wool panel (CW), manufactured by CEWOOD[®]. The mechanical properties of HMC and WMC showed that the bending strength difference was about 30%, with a better result for HMC. Compression strength was better for WMC by about 60% compared to that of HMC. The mechanical strength of HMC and HC materials was equal; both materials contained hemp shives but differed by the binding material. Water absorption and volumetric swelling tests showed that HMC and WMC could be considered as potential biosorbents. Ash content and elemental analysis showed that reference materials (CW, HC) contained significant amounts of inorganic compounds that decreased the biodegradation rate, compared to the case of HMC and WMC materials. The biodegradation results of HMC and WMC, after 12 weeks, revealed a mass loss (ML) above 70%, while in the case of EV, HC and CW, it was about 60%, 17% and only 6%, respectively. MBB were completely biodegradable.

Keywords: Mycelium-based biocomposites (MBB); mechanical properties; water absorption; biodegradation

1 Introduction

The idea of ecological materials is becoming more and more popular in society, which increases the challenges of our society to the transition towards a sustainable economy [1]. For this purpose, the use of non-renewable resources has to be reduced for the production of materials and consumer products [2]. Renewable mycelium-based materials have the potential to contribute to the new economy by replacing petroleum-based products such as plastics [2]. These bio-based products of mycelium-based materials could be used for thermal and acoustic insulation [3,4], and packaging [5].



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The mycelium-based materials have been produced mainly from mushroom forming fungi, which are known for their ability to colonize large areas in nature [2,6]. As an example are single individuals of the genus *Armillaria* able to colonize over 1000 ha of soil, making them the largest organisms on earth [7]. Furthermore, mushroom forming fungi are known for their ability to degrade lignocellulosic waste streams such as sawdust and straw. Fungi are organisms, which are able to give cohesion to incoherent materials due to the production of a mass of microscopic filaments, hyphae, which form the mycelium. Besides, fungi never produce real tissues, but still hyphae can display different types and specializations related to the substrate degradation and the development of reproductive structures. As a result, by biodegradation, the organic material is replaced by fungal biomass and within the substrate particles. At a certain moment, hyphae grow out of the substrate into the air creating a fluffy or compact layer covering the substrate. This compact layer is also known as fungal skin [2,6,8].

Fungal materials are distinguished as pure and composite fungal materials. The pure mycelium material's properties depend on the substrate, the type of fungus, and its growth conditions as well as post processing [9]. Using different mycelium material grown on cellulose, different properties of the material are improved. For example, the *Pleurotus ostreatus* mycelium on cellulose is stiffer compared to the *Ganoderma lucidum* mycelium, while addition of dextrose to the cellulose based substrate makes both fungal materials more elastic [10], and even a single gene can affect the material properties of the mycelium. The growth conditions provided affect the mycelial properties [2]. The composite fungal material's properties are determined by both the base of the organic material and the choice of mycelium, as the organic material will be more cellulose-containing, the easier it is to degrade. The white rot fungus *Trametes versicolor* is capable of degrading such complex compounds as lignin, as well as certain industrial contaminants [11].

So far, composite materials have been shown to exhibit properties similar to expanded polystyrene or other foams [3,4,12]. The main factors affecting the production of mycelium composites, and consequently, their mechanical behavior are the matrix (mycelium species), the lignocellulosic substrate, the interaction between fungi and their lignocellulosic substrate, and the process variables during manufacturing [13]. Studies show that, the same as for pure mycelium, the properties of the mycelium composites depend on the fungus, substrate, growth conditions, and processing of the material, as well as its additives. For instance, the different combination of the lignocellulosic substrate and fungi affects material density, mechanical strength, acoustic properties and absorption [2,14,15].

Mycelium composites are already commercially manufactured as interior decorative and packing materials [5,16] as well as acoustic wall panels [17]. The aim of this study was to develop MBB from the local agricultural and forestry waste-hardwood chips and hemp shives, and the mycelium of the white rot fungus *Trametes versicolor*. The material properties such as mechanical strength, water absorption, volumetric swelling, biodegradation, ash content and elemental composition were determined. The obtained mycelium composite materials were analyzed and compared with a commercial composite material from Ecovative Design[®] [16], an inorganic binder composite material of hemp shives and magnesium oxychloride (HC) [18,19], and a commercial cemented wood wool panel (CW), consisting of wood chips and a cement binder [20].

2 Methodology

2.1 Materials

For manufacturing of novel mycelium-based biocomposites (MBB), an agricultural and forestry byproduct from local industries was obtained and bounded together with a naturally growing fungal mycelium. The mycelium of the lignocellulosic basidiomycete *Trametes versicolor* was cultivated in controlled laboratory conditions at $22 \pm 2^{\circ}$ C and $70 \pm 5\%$ relative humidity. The fungal inoculum was spread on moistened and sterilized substrates, containing (a) hardwood chips and (b) hemp shives (Fig. 1).



Figure 1: Mycelium-based biocomposites (MBB) with hardwood chips (a) and hemp shives (b)

After the mycelium had fully overgrown the substrates, the material was transferred to molds 16 cm \times 4 cm \times 4 cm in size to continue the cultivation and obtain the shape of final products. Subsequent to the cultivation, the specimens were withdrawn from molds and dried at 93°C to deactivate the fungal growth. Two types of specimens–hemp mycelium composite (HMC) and wood mycelium composites (WMC)–were successfully obtained for further investigations (Fig. 2).



Figure 2: Material specimens of hemp mycelium composite (HMC) and wood mycelium composites (WMC)

The reference materials with the hemp organic substance is Ecovative[®] mycelium composite (EV) and hemp concrete (HC), obtained from hemp shives and a magnesium oxychloride binder. Also the reference material with wood chips is cemented wood wool (CW) from CEWOOD[®], which consists of wood chips and a cement binder. Those reference materials were prepared to compare the physical, mechanical and biodegradation properties with those of MBB. For this, the EV growth material was purchased from Ecovative Design LLC (Green Island, NY, USA) and the specimens were fabricated in the laboratory according to the growth instructions [16].

The HC material was obtained in a previous experimental research [18,19]. HC is a bio-composites material from hemp shives with a binder of magnesium oxide and magnesium chloride; the current specimen's material filler and binder ratio is 0.71, and density is 333 kg/m³ [18]. The EV material made in the laboratory according the manufacturer's instructions is with a density of 120 kg/m³. The commercial CW fibrolite material (CEWOOD[®], Ltd.,) [20] was purchased from the retail. CW is a cemented wood wool panel with a density of 460 kg/m³, widely used as insulation in wall constructions and as finishing material with a good acoustic ability.

2.2 Methods

In this research, novel MBB material specimens, which were obtained from two types of substrates from hemp shives and hardwood chips, as HMC and WMC, were tested to determine the mechanical properties of

the material, which is a tool for the potential application of the material. The obtained material–HMC and WMC–was tested for material bending strength (BS), compression strength (CS), water absorption and biodegradation.

2.2.1 Mechanical Strength

Tests of the BS of conditioned HMC, WMC and EV materials-were performed on a Zwick Z100 universal testing machine according to DIN 52186 (3-point flexure test) until collapse for the BS on the pre-prepared materials by dimensions of 16 cm \times 4 cm \times 4 cm. It should be noted that all edges of the material are not regular, and smooth, which was influenced by the experimental design of the material.

Tests of the CS of conditioned HMC, WMC and EV were performed according to EN 826 on an Instron E3000 universal testing machine for more accurate measurements—to the defined deformation of 14 mm of the material at a speed of 2 mm/s. The tested material was pre-prepared with average dimensions of 40 mm \times 40 mm; a smooth and even shape form was not obtained at all edges by cutting the specimens.

2.2.2 Water Absorption and Volumetric Swelling

The water absorption and thickness swelling of different composites were measured according to ASTM D1037: 2012, with modifications using 3 cm \times 3 cm specimens (6 replicates were used in each group). The specimens were immersed in distilled water and the weights and volumes were measured after 2 h and 24 h. The water absorption and volumetric swelling values were determined from the weight and volume difference in relation to the initial weight and volume.

2.2.3 Biodegradation

The specimens of HMC, WMC and the reference materials (EV, HC, CW) with dimensions of 8 cm \times 4 cm \times 4 cm were composted in a grass and hardwood sawdust containing compost pile. A specially designed inoculum with four bacteria strains (*Pseudomonas* spp., *Nitrosomonas* spp., *Nitrobacter* spp. and *Sarcina* spp.) isolated from a biologically active sludge and two fungal strains (*Trichoderma* spp.) was added to facilitate the composting process [21]. The composting was carried out in the open air in the summer-autumn season of 2019 at intervals of 4, 10 and 12 weeks to observe the biodegradability of materials. The composting parameters –pH and temperature–were measured regularly (Fig. 3).



Figure 3: Temperature of the compost site and outside air during composting

The specimens were placed in a compost pile in the thermophilic stage of composting when the temperature in the pile reached 60°C. During 12 weeks of composting, aeration was performed twice in the compost site, which resulted in a sharp drop in composting temperature.

Subsequent to composting, the specimens were removed from the composting site, brushed free of compost substances and dried at room temperature until the constant weight. The percentage mass loss (ML) of the specimens was the measure for the extent of biodegradation.

2.2.4 Ash Content

Ash content was determined according to TAPPI T 210: 2003. 2 g of specimen was weighted into a crucible and placed in a Carbolite furnace ELF 11/6B at $575^{\circ}C \pm 25^{\circ}C$ for 3 h. After ignition, the crucible was placed in a desiccator to cool to room temperature, weighted, and the percentage of ash was calculated based on the moisture-free weight of the specimen.

2.2.5 Elemental Analysis

For elemental analysis (C, H, N and S) according to LVS CEN/TS 15104:2011, homogenized specimens (30 mg) were packed in a tin foil, weighed, placed into the carousel of an automatic specimen feeder and analyzed with an Elementar Analysensysteme GmbH (Germany) Vario MACRO CHNS with a combustion tube temperature of 1150°C. The original matrix of the specimen was destroyed under these conditions through subsequent catalytic reactions.

3 Results and Discussion

HMC with a density of 134 kg/m³ and WMC with a density of 179 kg/m³ were obtained. The Ecovative[®] (EV) was fabricated in the laboratory according to the growth instructions, with a density of 120 kg/m³.

3.1 Mechanical Strength

The BS test showed better results for HMC compared to the case of WMC and EV (Tab. 1). The results of HMC and WMC were closer compared with the equal EV, as well as the result compare HMC and EV is 5 times different, which were equal from viewpoint of the used substrate in the mycelium composite–hemp shives. The large difference was due to the granulometric size of the substrate used; larger size granulometric hemp shives were used in HMC.

Table 1: Mechanical strength of the tested specimens-hemp mycelium composite (HMC), wood mycelium composites (WMC), Ecovative[®] mycelium composite (EV), hemp concrete (HC) and cemented wood wool (CW)

Mechanical strength	HMC	WMC	EV	CW [20]	HC [18,19]
Bending strength (MPa)	0.16 ± 0.02	0.11 ± 0.01	0.05 ± 0.01	1.3	0.16
Compression strength (MPa)	0.36 ± 0.05	0.52 ± 0.08	0.23 ± 0.02	0.3	0.36
Density (kg/m ³)	134	179	120	460	333

The given results, comparing our material with the HC material with a density of 333 kg/m³ and BS of 0.16 MPa [18,19], showed that our material–HMC, which was based on hemp shives, was with equal BS. The higher the filler to binder ratio for HC in these particular specimens, which was 0.71 [19], the lower the result of the HC BS, but better using a thermal insulation layer in construction, for example, HC with a filler to binder ratio of 0.5 obtained at a BS of 0.35 MPa was better as a load bearing layer for constructions [19]. The BS of the CW material, comparing to that of the WMC material, was near 10 times bigger; for the CW material, BS is \geq 1.3 MPa (according to manufacturer's data) [20]. HMC and WMC were not comparable to structural building materials; those could be considered as a heat insulating material according to BS results.

The test of CS with an Instron E3000 universal testing machine showed a better result for the WMC material compared with the case of HMC and EV (Tab. 1). The results of each material were different, the closest results were for HMC and EV specimens; these two material specimens contained hemp shives. It should be noted that the shapes of the materials' specimens were irregular, which was made by the material filler–for example, HMC and EC contained hemp shives which were larger in the granulometric size compared to hardwood chips. Hardwood chips contained 51% particles with size of < 10 mm, 46% of 10–20 mm and 3% of > 20 mm, while hemp shives contained 49% particles with size of < 10 mm, 21% of 10–20 mm and 16% of > 40 mm.

CS was better for WMC than for CW specimens. Comparing WMC with similar experimental research of mycelium composites, as Mycelium brick [6] with a density of 318 kg/m³, the results were similar to those for WMC with a CS of 0.52 MPa, and Mycelium brick with 0.5 MPa [6]; for structural building materials such as clay brick, CS is 8.6 MPa–17.2 MPa [6].

However, the result of HMC and HC (333 kg/m³) [18,19] was 0.36 MPa for both materials; equivalent results were obtained using both the organic binder as mycelium and the inorganic binder as magnesium oxychloride, which was used in the particular HC material as the binder. The same hemp shives were used in both materials; the binder used did not affect the CS. The HC layer is used in the wall structure as insulation material [18].

WMC, comparing in terms of CS with the CW with a CS of ≥ 0.3 MPa (according to manufacturer's data), was with an equal value [20]. However, after the thermal conductivity test, WMC and HMC materials showed that they are suitable as an insulation material; for example, the polystyrene insulation material's CS is 0.03 MPa–0.054 MPa (manufacturer's data) [22]; the same is for rock wool insulation boards – CS ≥ 0.03 MPa (manufacturer's data) [23]. For example, thermal conductivity for polystyrene is 0.033 W/mK and for rock wool ≥ 0.035 W/mK, but for Mogu[®] acoustics wall panel modules (180 kg/m³), which are made from soft foam-like mycelium materials and upcycled textile residues, the thermal conductivity is 0.05 W/mK, BS is 0.05 MPa and CS, according to the EN 826 test, is 0.01 MPa [17]. Consequently, based on the thermal properties of the existing commercial material, equivalent thermal values can be predicted for HMC and WMC materials, so it is necessary to perform a thermal conductivity test to confirm this.

3.2 Water Absorption and Volumetric Swelling

The water absorption and swelling are important properties for the application of mycelium-based composites as indoor particle boards or insulation elements, and they characterize the durability of the material. Fig. 4. shows the water absorption and volumetric swelling results of all the tested specimens. HMC and WMC have the highest water absorption, reaching 400%-550% after 24 h. The lowest water absorption is for CW (~30%) and HC (~65%), probably, due to the use of magnesium oxychloride concrete and cement for fabrication of these specimens. The WMC, HC and CW specimens showed no significant difference in water absorption after 2 h and 24 h. Water absorption in similar research of hybrid panel composites based on wood, fungal mycelium, and cellulose nanofbrils after 24 h of the immersion period was 120%–240% [24]. These results are comparable with EV which is made of woodmycelium particles. Despite the similar density and preparation method, HMC and WMC had 2-4 times higher water absorption. Probably, wood particles in WMC and hemp in HMC are too hydrophilic and need additional treatment before the preparation of the composite to limit water absorption. Myceliumbased composites with T. versicolor and five different types of fibres (hemp, flax, flax waste, softwood, straw) were tested for water absorption rate. Water absorption, after 24 h for chopped hemp, flax and straw composites, was 24.5%, 30.3% and 26.8%, respectively. The growth on hemp specimens resulted in a denser outer hydrophobic mycelial layer, compared to flax and straw, explaining the lower water absorption rate of hemp [24].



Figure 4: Water absorption (left) and volumetric swelling (right) after 2 h and 24 h of the tested specimenshemp mycelium composite (HMC), wood mycelium composites (WMC), Ecovative[®] mycelium composite (EV), hemp concrete (HC) and cemented wood wool (CW)

Volumetric swelling of all tested specimens did not exceed 5% and was similar after 24 h, considering the error values. This indicates that the increased water absorption of HMC, WMC and EV affected the material's dimensions negligibly. This can be attributed to the low density of these specimens and a large surface in material pores for water absorption is available. Probably, water related properties depend on the composite manufacturing methodology, because the thickness swelling of hybrid panel composites based on wood, fungal mycelium, and cellulose nanofbrils after 24 h can reach up to 50%–130% [24].

3.3 Biodegradation

The specimen ML after composting is shown in Fig. 5. HMC and WMC specimens, within 12 weeks, were fully degraded in the compost environment having ML over 70%. EV specimens also were severely degraded with ML 60%. A higher biodegradation ability was observed for WMC specimens than for the hemp containing HMC and EV. WMC specimens, after 10 and 12 weeks, were decomposed in separate particles and it was difficult to collect a sound specimen to estimate a correct ML.



Figure 5: Biodegradation in compost environment during 12 weeks of the tested specimens-cemented wood wool (CW), hemp concrete (HC), Ecovative[®] mycelium composite (EV), wood mycelium composites (WMC) and hemp mycelium composite (HMC)

The degradation of HC specimens was limited. After 4 weeks, even an increased ML was observed, most likely because of the water absorption by concrete and slow drying afterwards. However, after 12 weeks, a partial decomposition with the ML of 17% occurred (Fig. 6). Obviously, hemp shives were degraded from the outer layers of specimens while the concrete barrier prevented the distribution of composting microorganisms into the HC specimens. Another reason for the slow degradation of HC can



Figure 6: Material specimens (HMC, WMC, EV, CW, HC) after 12 weeks of biodegradation test

be attributed to the alkaline reaction of concrete that is a growth limiting factor for lignocellulose degrading fungi. The lowest degradation ability was for CW specimens with a ML of 6% at the end of the composting experiment. The reasons for the restricted biodegradation could be similar to those of HC.

3.4 Ash Content and Elemental Composition

The ash content and elemental composition of the tested materials are shown in Tab. 2. The obtained results showed that CW and HC contain significant amounts of inorganic compounds. The organic part of these composites contained relatively little nitrogen compounds and can predict their poor biodegradability. The composition of CW did not change significantly, as shown by the composition of the elements before and after composting. The ML of the HC specimen reached 17% and, at the same time, the composition change of the specimen could be observed. The carbon and nitrogen content increased after composting, possibly due to the carbohydrate decomposition, but further analyses are necessary to confirm this assumption. The other specimens initially decomposed at the same rate, although their composition differed significantly. The EV contained relatively large amounts of nitrogen and the C/N ratio was favourable for biodegradation [25], but this specimen decomposed more slowly

Specimens	Ash (%)	N (%)	C (%)	Н (%)	C/N ratio				
Before composting									
CW	64.3	0.15 ± 0.01	18.1 ± 0.1	2.8 ± 0.1	118				
HC	48.1	0.26 ± 0.03	17.0 ± 0.6	2.8 ± 0.1	64				
EV	5.0	1.49 ± 0.12	45.9 ± 0.1	4.5 ± 0.1	31				
WMC	0.6	0.67 ± 0.04	48.3 ± 0.2	4.2 ± 0.2	72				
HMC	4.6	1.55 ± 0.05	46.2 ± 0.2	4.3 ± 0.1	30				
After composting									
CW	64.1	0.15 ± 0.01	18.1 ± 0.1	2.3 ± 0.2	117				
HC	36.9	0.42 ± 0.01	25.4 ± 0.2	3.5 ± 0.3	61				
EV	6.5	1.94 ± 0.12	48.4 ± 0.3	4.3 ± 0.1	25				
WMC	1.4	0.92 ± 0.06	49.1 ± 0.1	4.3 ± 0.2	54				
HMC	4.6	1.49 ± 0.01	48.4 ± 0.2	4.3 ± 0.1	33				

Table 2: Ash content and elemental composition of the studied materials before and after biodegradation in the compost environment

than WMC and HMC. Wood is generally more resistant to biodegradation because of its high lignin and low nitrogen content (C/N ratio 72), so, the lignin-degrading microorganisms were added to the compost substrate.

4 Conclusions

Mechanical strength tests indicated that the BS of HMC was higher by \sim 30% compared to that of WMC, while CS was higher by \sim 60% for WMC compared to that of HMC. The HMC and HC had equal mechanical strength in terms of BS and CS. Probably, this is because both materials were prepared using hemp shives, but different binder material was used.

The water absorption and volumetric swelling tests showed that HMC and WMC could be considered as biosorption materials due to the increased water uptake, which is 2–4 times higher compared to the similar commercial product EV. The lowest water absorption was observed for CW (\sim 30%) and HC (\sim 65%), probably, due to the use of inorganic magnesium oxychloride concrete and cement for production of these materials. The increased water absorption of HMC, WMC and EV affected the material dimensions negligibly, and volumetric swelling did not exceed 5%.

Ash content and elemental analysis showed that the reference materials (CW and HC) contained significant amounts of inorganic compounds that affected the rate of biodegradation compared to the case of HMC and WMC. The biodegradation test showed that the ML of HMC and WMC after 12 weeks of biodegradation was above 70%, while for EV, HC and CW, it was 60%, 17% and 6%, respectively. Our MBB were completely biodegradable.

Based on the results obtained, further studies are necessary to evaluate the material as a thermal insulation or sorbent material.

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