Review | Received 7 November 2025; Revised 10 February 2026; Accepted 25 February 2026; Published 10 March 2026
https://doi.org/10.55092/bm20260003

Heat pressing of mycelium-based composites: an integrative review of processing parameters and mechanical properties

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Highlights:

- Heat pressing activates lignin plasticization and Maillard cross-linking while preserving chitin integrity (stable to ~300 ℃).
- Pressure is the dominant processing parameter; 30%–50% moisture content optimizes lignin flow and interfacial bonding.
- Optimized processing achieves 37.6 MPa flexural strength, comparable to commercial medium-density fiberboard.
- Cross-study variability demands adoption of standardized testing protocols for field advancement.

Abstract: Mycelium materials are an emerging and promising class of biomaterials currently used primarily for biodegradable packaging and insulation. Their mechanical properties limit them to non-structural applications akin to polystyrene; however, post-processing techniques such as heat pressing may widen their possible applications, with some studies achieving metrics comparable to commercial particleboards. This integrative review analyzes studies focused on heat pressing mycelium composites, comparing the available quantitative data on mechanical properties. Among conventionally processed samples, mean tensile strength reached 6.3 MPa and elastic modulus 2138 MPa, with considerable variability across studies. Recent advances have achieved exceptional results: ultra-high-pressure processing (100 MPa) yielded tensile strengths of 12.5 MPa, approaching values for engineering plastics, while semi-wet hot-pressing achieved flexural strengths of 37.6 MPa. Correlation analysis revealed that pressure exerted a stronger influence on mechanical properties than temperature, with moderate to strong positive correlations across all measured outcomes. Optimal material properties appear achievable through temperatures of 130–170 ℃ combined with elevated pressures and controlled moisture content (~30%), which facilitates lignin plasticization and enhanced bonding. However, substantial heterogeneity in experimental methods and inconsistent property reporting across studies complicate direct comparisons. This review highlights the urgent need for standardized manufacturing and testing protocols in mycelium composite research and demonstrates the potential of heat pressing to produce sustainable, biodegradable alternatives to conventional particleboards.

Keywords: mycelium composites; heat pressing; sustainable materials; biomaterials

1. Introduction

1.1. Problem statement

The building materials and packaging industries face mounting pressure to reduce their environmental footprint. Conventional particleboards and medium-density fiberboard (MDF) rely on petroleum-derived adhesives, predominantly urea-formaldehyde resins, which emit volatile organic compounds throughout their service life and persist in landfills for decades [1]. Expanded polystyrene (EPS), ubiquitous in protective packaging, derives entirely from fossil fuels and accounts for a substantial fraction of marine plastic pollution due to its fragmentation into microplastics. These environmental burdens have intensified demand for biodegradable, carbon-neutral alternatives that can match the mechanical performance of incumbent materials.

Mycelium-based composites have emerged as a compelling candidate to address this need. These materials are produced through the natural growth of fungal mycelium within lignocellulosic or other organic substrates, resulting in composites that are lightweight and fully biodegradable, with significantly reduced carbon footprints compared to traditional materials [2]. Unlike synthetic adhesives, the fungal mycelium acts as a self-assembling biological binder, requiring no external energy input during growth and utilizing agricultural waste streams—such as sawdust, straw, and hemp shives—that would otherwise be burned or landfilled [3]. As they may utilize exclusively renewable or natural ingredients in their composition, mycelium composites contribute to a circular economy by sequestering atmospheric carbon dioxide during fungal growth and returning nutrients to the soil upon decomposition.

However, realizing this environmental promise requires achieving mechanical properties sufficient for practical applications. The critical performance thresholds are well-established: commercial particleboard typically exhibits tensile strengths of 0.3–0.5 MPa and elastic moduli of 1500–3000 MPa, while MDF achieves flexural strengths of 20–40 MPa [4]. Expanded polystyrene, though mechanically weak (compressive strength 0.07–0.3 MPa), offers consistent properties and excellent thermal insulation. For mycelium composites to displace these materials, they must approach or exceed these benchmarks while retaining their biodegradability and low embodied energy.

Historically, mycelium-based materials were categorized primarily as low-density foams (30–200 kg/m^3), utilized for non-structural applications such as biodegradable packaging, acoustic panels, and thermal insulation [1]. These “first-generation” materials relied on natural hyphal growth to bind agricultural waste substrates into a cohesive, albeit soft, shape. Despite their environmental advantages, current applications of mycelium composites remain predominantly limited to non-loadbearing uses due to several inherent challenges: relatively low mechanical strength, sensitivity to moisture, and variability in material properties arising from their biological nature [5]. The geometric and mechanical limitations of foam-like structures—characterized by low tensile strength (< 0.1 MPa), high porosity, and significant water susceptibility—have hindered their adoption in the construction and automotive sectors where they could deliver the greatest environmental benefit [6].

Heat pressing has demonstrated potential in bridging this performance gap. By subjecting the porous mycelium matrix to simultaneous heat and pressure, the material undergoes a fundamental phase change, transitioning from a viscoelastic foam to a rigid, high-density composite analogous to MDF or

engineered woods. The effectiveness of this technique lies in its ability to facilitate stronger bonding between the mycelium's chitin and the substrate's lignin polymer chains through thermal crosslinking, while simultaneously achieving greater material density [5]. Figure 1 illustrates this application spectrum, showing how increasing processing intensity—from simple drying through conventional pressing to advanced high-pressure techniques—progressively expands the range of viable applications from low-strength packaging toward load-bearing structural components. If heat-pressed mycelium composites can consistently achieve tensile strengths exceeding 5 MPa and flexural strengths approaching 20 MPa, they would become viable replacements for conventional particleboards in furniture, interior paneling, and construction applications, potentially displacing millions of tons of formaldehyde-emitting, non-biodegradable materials annually.

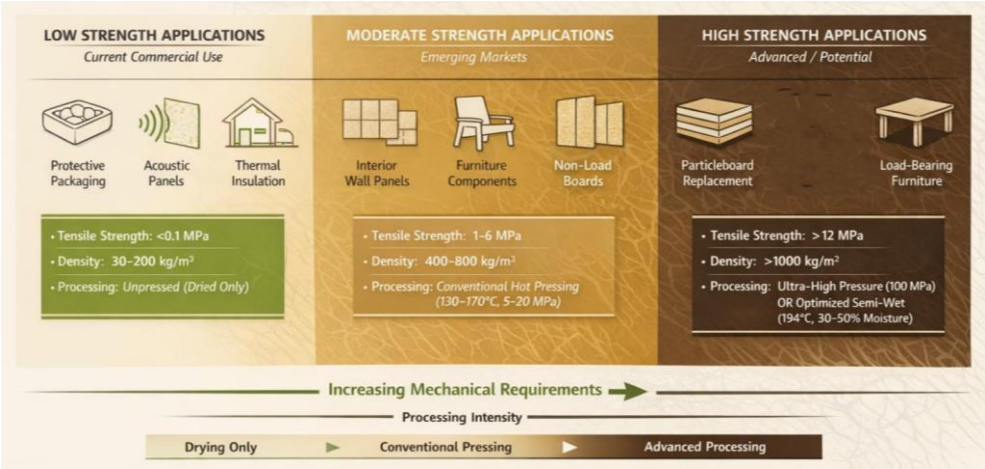

Figure 1. Application spectrum of mycelium composites. Figure partially generated with AI.

1.2. Research objectives

The research landscape in mycelium composites has evolved rapidly over the past decade [3]. Initial studies focused primarily on proving the concept and basic characterization. Recent years have seen a shift towards optimizing growth conditions, exploring diverse substrates, and enhancing material properties through post-processing techniques like heat pressing [7]. Most recently, research has advanced to fundamental alterations of the material's micro-architecture through ultra-high-pressure processing and interface engineering [6]. This evolution underscores the need for a comprehensive review of the current state of knowledge.

This integrative review analyzes studies focused on heat pressing mycelium composites, comparing the available quantitative data on mechanical properties. By aggregating and analyzing these findings, we aim to identify correlations, trends, gaps, and potential areas for further research, contributing to our incipient understanding of heat-pressed mycelium materials.

1.3. Overview of mycelium composites

The production of mycelium composites lies at an intersection of biology and materials science, leveraging the natural growth characteristics of filamentous fungi. These organisms have evolved sophisticated mechanisms for colonizing and breaking down organic materials, particularly lignocellulosic substances. During the growth phase, fungal hyphae, which are thread-like cellular structures, penetrate and weave

through the substrate material while secreting various enzymes [8]. These enzymes partially degrade the substrate components, particularly lignin, cellulose and hemicellulose, which the fungi utilize for nutrition. Simultaneously, the growing hyphae produce new biomass, primarily composed of chitin and other polysaccharides, creating an intricate three-dimensional network throughout the substrate [5]. This creates two key binding mechanisms:

(1) Mechanical Interlocking: The branching and growth patterns of fungal hyphae create a complex network that physically entangles with substrate particles. This is evidenced by scanning electron microscope (SEM) imaging showing hyphal networks extending through and around discrete particles [5].

(2) Chemical Bonding: The fungal cell walls contain chitin, chitosan and β-glucans that can vary in their compositional ratio depending on the species [9]. The chitin in fungal cell walls is a strong material, with a Young's modulus (or elastic modulus, *i.e.*, the ability to resist deformation under stress) of up to 3.7 GPa [4]. As such, the strength of these composite materials is highly dependent on the fungal biomass, and chemical interactions are confirmed through FTIR analysis showing hydrogen bonding with it [10]. Moreover, during growth and subsequent processing, various chemical interactions occur between fungal cell wall components and substrate materials, binding them together [3].

Figure 2 illustrates the simplified production process for heat-pressed mycelium composite boards, from substrate preparation and fungal inoculation (usually with grain spawn) through colonization, deactivation, pressing, and finishing.

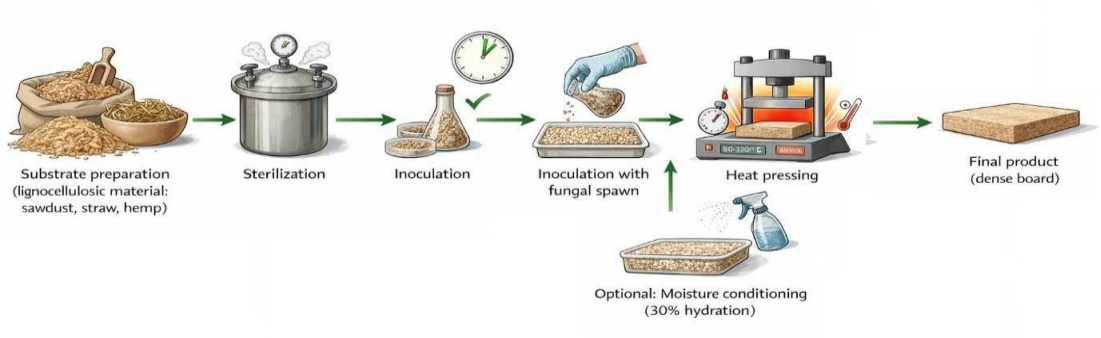

Figure 2. Simplified Process for heat-pressed mycelium composite board. Figure partially generated with AI.

1.4. Heat pressing

1.4.1. Thermochemistry of the mycelium-lignocellulosic interface

To fully appreciate the effects of heat pressing, one must understand the thermodynamic behaviors of the composite's constituent biopolymers. A mycelium composite is a multi-phase system comprising the fungal cell wall (chitin, β-glucans, proteins), the substrate (cellulose, hemicellulose, lignin), and water. The transformation from a loose aggregate to a solid board is governed by the glass transition temperatures (Tg) of these components and the chemical cross-linking reactions activated by thermal energy.

In conventional wood composites, synthetic resins like urea-formaldehyde are added to bind fibers. In mycelium composites, the substrate itself provides the binder in the form of lignin. Lignin is a complex, amorphous polyphenolic polymer that acts as the "glue" in plant cell walls. Lignin acts as a thermoplastic; it is rigid and glassy at room temperature but softens and flows when heated above its

glass transition temperature (Tg) [11]. The Tg of "native" lignin is typically between 127 ℃ and 193 ℃ in a dry state, depending on the source [12]. However, processing dry lignin requires temperatures that often induce thermal degradation of other components.

The presence of moisture is therefore catalytic. Water molecules diffuse into the amorphous regions of the lignin polymer network, disrupting inter-chain hydrogen bonds and increasing free volume. This phenomenon, known as plasticization, significantly depresses the Tg [13]. Sorption of water by lignin causes a pronounced decrease in the softening temperature—in some cases to as low as 54 ℃ [12]. The glass transition temperature of lignin in moist wood is approximately 80–100 ℃ [11,14]:

- Dry State (< 5% moisture): Tg ≈ 140 ℃. Processing requires T > 160 ℃ to achieve adequate flow, risking charring.
- Hydrated State (10%–15% moisture): Tg drops to ≈ 80–100 ℃.
- Optimal State (30% moisture): At 30% hydration, the lignin becomes sufficiently mobile at 130–150 ℃ to flow around the fungal hyphae, effectively "wetting" the chitinous reinforcement [15].

This mechanism explains why Liu *et al.* [15] observed superior mechanical properties in samples pressed at 30% hydration compared to dry samples. The "wet" lignin could flow and redistribute stresses, whereas the "dry" lignin remained brittle and glassy, failing to form a continuous matrix. Additionally, moist lignin becomes tacky with increased temperatures, enhancing the bond strength between lignin and fibers in close contact [16]. The "steam shock" effect created by the moisture also facilitates rapid heat transfer into the core of the composite, allowing for better compaction [17].

Recent work by Song *et al.* [18] has formalized this moisture-dependent processing as "semi-wet hot-pressing," where material moisture content is controlled between 30%–50% prior to pressing. This approach represents a middle ground between conventional wet processing (65%–70% moisture, which requires extensive water resources) and dry processing (6%–8% moisture, which typically necessitates synthetic adhesives). Under semi-wet conditions, fibers are dispersed and transported in water, facilitating hydrogen bond formation and improving inter-fiber adhesion. Simultaneously, mycelial proteins and polysaccharides undergo denaturation and softening at elevated temperatures, promoting compaction and densification. Using response surface methodology to optimize this process, Song *et al.* achieved a modulus of rupture of 37.64 MPa, demonstrating the critical role of controlled moisture content in maximizing mechanical performance.

Figure 3 illustrates the relationship between moisture content and the glass transition temperature (Tg) of lignin. While intact native lignin within the wood structure exhibits a dry Tg near 200 ℃, the 140 ℃ baseline plotted here reflects values typical for isolated lignins. As Back and Salmén [13] explicitly note, the extraction of lignin can "significantly alter its chemical and physical structure" and reduce its molecular weight, which lowers the glass transition temperature. This lower baseline represents the state of lignin in mycelium composites, which undergoes structural alteration and depolymerization via fungal enzymes during colonization. As hydration increases, this Tg is progressively depressed through plasticization, significantly widening the practical hot-pressing window.

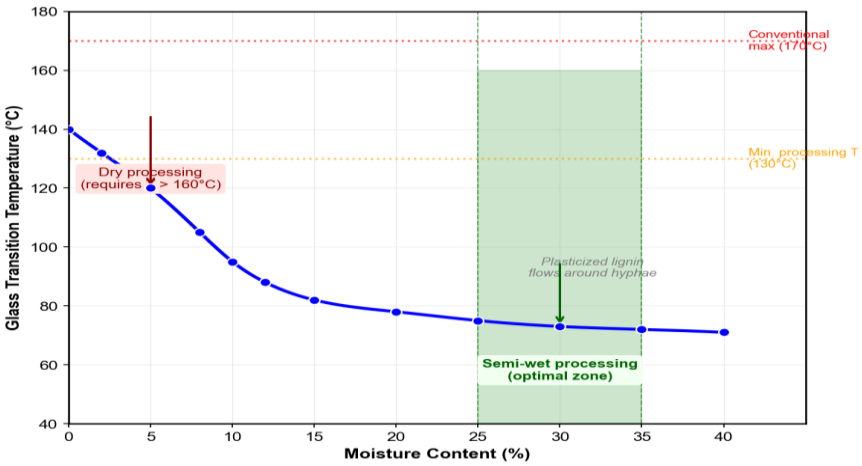

Figure 3. Effect of moisture content on lignin glass transition temperature.

1.4.2. Thermal stability of chitin

While lignin provides the matrix, the fungal hyphae act as the reinforcement fibers. The cell wall of Basidiomycetes (the class of fungi most commonly used, e.g., Ganoderma, Trametes, Pleurotus) is a complex composite of crystalline chitin nanofibers embedded in a matrix of β-glucans and glycoproteins [19]. Chitin is a linear polysaccharide composed of β-(1,4)-linked N-acetylglucosamine units [20]. It is structurally similar to cellulose but possesses acetamide functional groups that allow for hydrogen bonding.

The α-chitin polymorph found in fungal walls is highly crystalline and remarkably thermally stable. Thermal gravimetric analysis (TGA) has demonstrated that thermal degradation of solid chitin of various origins occurs only above 300 ℃ [21]. In situ optical microscopy studies have provided direct evidence that chitin and fungal cells with chitinous cell walls remain undegraded at temperatures well over 200 ℃ under hydrothermal conditions [21]. Indeed, the complete dissolution of fungal hyphae (Flammulina velutipes) and chitin was observed at almost the same temperature (380–390 ℃), strongly suggesting that chitin that makes up fungal cell walls is robust and remains intact up to approximately 380 ℃ [21]. In fungal cell walls, chitin exists as fine bundles, 20–30 nm in width [19].

This creates a defined thermodynamic processing window for heat pressing. The temperature must be high enough to soften the lignin (> 130 ℃) but low enough to preserve the crystalline integrity of the chitin. While chitin itself is stable to very high temperatures, the degradation of other fungal cell wall components such as glucans and chitosan begins in the temperature range of 250–350 ℃ [22]. More critically, the degradation of hemicellulose in the substrate and the potential for bond cleavage in the overall composite matrix begins at lower temperatures. When temperatures exceed 180 ℃, the covalent bonds formed at lower temperatures begin to break down [23], and surpassing 200 ℃ risks severe thermal degradation of hemicellulose and other biological components [1]. Therefore, the practical processing window of 130–170 ℃ represents a balance between achieving adequate lignin flow and preserving the structural integrity of all composite components.

1.4.3. Chemical cross-linking: the maillard reaction

Beyond physical phase transitions, heat pressing induces irreversible chemical reactions that harden the composite. The most prominent of these is the Maillard reaction, often evidenced by the darkening or

"browning" of the material surface during pressing. The Maillard reaction was first reported by French chemist Louis Maillard in 1912 [24] and is a complex series of non-enzymatic browning reactions between reducing sugars (derived from the thermal hydrolysis of hemicellulose or residual culture media) and amino groups (present in fungal proteins and the deacetylated units of chitin/chitosan) [25,26].

Recent research suggests the Maillard reaction produces reactive α-dicarbonyl intermediates that crosslink proteins, forming high-molecular-weight melanoidin complexes [27]. This crosslinking effectively bonds the fungal cell wall proteins to the plant polysaccharides, increasing the stiffness (Young's Modulus) of the interface and reducing water sensitivity [25]. The Maillard reaction has been shown to effect protein crosslinking on timescales relevant to industrial processing [28]. Chen *et al.* [6] confirmed that mycelium grown in nitrogen-rich liquid cultures (higher protein content) exhibited more intense browning and higher stiffness upon pressing, attributing this directly to enhanced Maillard cross-linking.

1.4.4. Processing parameters

The temperature selection during heat pressing is critical, as it governs both chemical reactions and physical transformations. The optimal processing range of 130–170 ℃ provides sufficient thermal energy for effective crosslinking while avoiding excessive degradation of biological components. When temperatures exceed 180 ℃, the covalent bonds formed at lower temperatures begin to break down [23], and surpassing 200 ℃ risks severe thermal degradation of biological components [1]. The applied pressure typically ranges from 2.5 to 20 MPa in conventional processing, though recent research has explored ultra-high pressures up to 100 MPa [6], working in concert with the heat to reshape the material's internal structure.

This combination of heat and pressure triggers several concurrent mechanisms that enhance the material's properties. The pressure physically compacts the composite, reducing porosity and increasing density, while the heat promotes crosslinking between fungal chitin and substrate lignin, creating stronger chemical bonds [10]. Together, these forces reorient fibers and create a more uniform, consolidated structure [8]. The effectiveness of this process is demonstrated by Yang *et al.*'s [10] finding that heat pressing a mixture of sawdust lignin and chitin from crab shell powder in a 9:1 ratio at 200 ℃ produced materials with triple the mechanical strength of pressed sawdust alone. Unlike traditional drying, which primarily serves to deactivate the fungus and remove moisture, heat pressing actively transforms both the physical structure and chemical composition of the material [29].

Figure 4 summarises the thermochemical processing window for mycelium composites, delineating the temperature thresholds for lignin softening, optimal cross-linking, and the onset of thermal degradation of composite components.

The optimization of processing parameters through statistical design of experiments has emerged as a powerful approach for maximizing composite performance. Song *et al.* [18] employed Box-Behnken response surface methodology to systematically investigate the interactions between pressure, temperature, and time in mycelium-bound boards. Their analysis revealed that pressure and temperature exhibited the most pronounced effects on board performance, with optimal conditions of 10 MPa, 194 ℃, and 31 minutes yielding exceptional results. This systematic optimization approach addresses the variability issues highlighted throughout the mycelium composite literature and provides a framework for industrial process development.

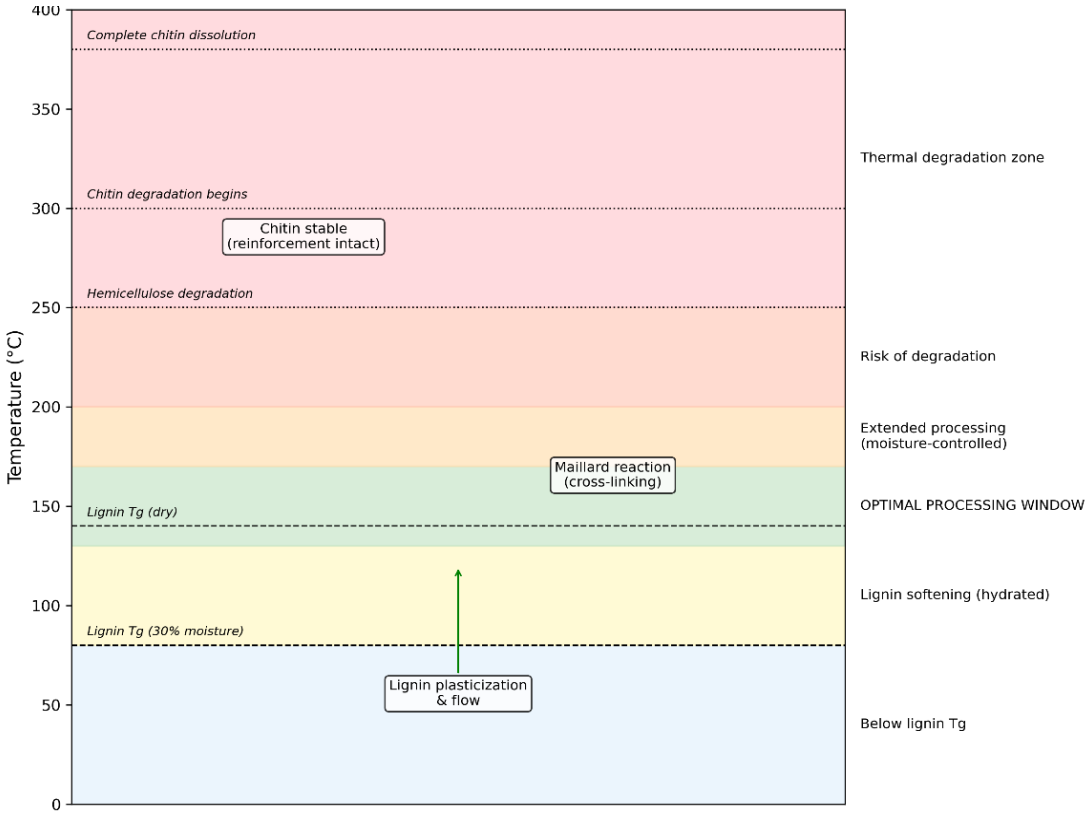

Figure 4. Thermochemical processing window for mycelium composites.

Recent studies have expanded the substrate and species diversity in mycelium composite research. Shakir *et al.* [30] demonstrated that rice husk waste, combined with *Rhizopus oligosporus* mycelium as a bio-binder, can produce composites with flexural strengths reaching 5.79 MPa through hot pressing at moderate temperatures (130 ℃). Similarly, spent mushroom substrates have emerged as promising feedstocks for formaldehyde-free bio-boards, with Khoo *et al.* [4] achieving internal bonding strengths of 2.51 MPa using *Ganoderma lucidum* substrates pressed at 160 ℃, and Yan *et al.* [31] reporting internal bonding strengths of 2.16 MPa with corn stalk/*Pleurotus ostreatus* substrate composites pressed at 190 ℃. These developments underscore the versatility of heat pressing across diverse fungal species and agricultural waste streams.

1.5. Biological interface formation

The efficacy of heat pressing is not merely a function of mechanical compaction but is predicated on the quality of the biological interface—the boundary zone where fungal hyphae intimately contact, penetrate, and chemically bond with substrate particles. In a mycelium composite, the fungus acts simultaneously as the reinforcement fiber (through its chitinous hyphal network) and as the agent of matrix modification (through enzymatic degradation of substrate lignin). Consequently, the biological preform—the colonized substrate prior to pressing—establishes the upper bound of achievable mechanical performance. This section consolidates recent findings on how the biological preform is engineered through species selection, growth regulation, nutritional optimization, and substrate geometry, and how these pre-pressing factors propagate through to determine the properties of the heat-pressed composite.

1.5.1. Mycelial growth regulation

Species Specificity and Hyphal Architecture. The selection of a fungal species serves as the foundational design decision, as it dictates the intrinsic architectural properties of the biological binder. Fungal mycelia are not uniform structures; they exhibit distinct hyphal systems that vary in tensile strength, elasticity, and growth kinetics. The monomitic system, characteristic of species such as Pleurotus ostreatus, consists solely of generative hyphae—thin-walled, septate structures rich in cytoplasm but relatively low in structural polysaccharides [9]. While P. ostreatus is favored for its aggressive colonization speed and ability to outcompete contaminants, the resulting preform is comparatively soft. Upon heat pressing, monomitic networks compact readily but may provide less rigid fibrous reinforcement compared to species with more complex hyphal systems.

In contrast, Ganoderma lucidum and Trametes versicolor exhibit dimitic or trimitic hyphal systems comprising up to three distinct hyphal types: generative hyphae (for reproduction and expansion), skeletal hyphae (thick-walled, unbranched, chitin-rich), and binding hyphae (highly branched structures that interweave the network) [19]. The skeletal hyphae act as microscopic reinforcement elements, providing tensile stiffness, while the binding hyphae function as cross-links that enhance toughness and resist delamination. Composites utilizing trimitic species may therefore retain superior mechanical properties after pressing because the skeletal hyphae resist thermal degradation and maintain their reinforcing function even as the surrounding matrix plasticizes—though this hypothesis has not been tested through controlled species comparisons under identical pressing conditions.

Beyond the traditional Basidiomycota, Rhizopus oligosporus—a Zygomycete used in food fermentation—has been explored as a candidate for high-throughput production [30]. Unlike the septate hyphae of basidiomycetes, Rhizopus forms coenocytic (non-septate) hyphae and can achieve substrate colonization in as few as 2–4 days, compared to 14–28 days for typical white-rot fungi. However, the resulting network is often less dense and lacks the robust “skin” associated with Ganoderma species. Composites made with Rhizopus thus rely more heavily on densification of the substrate particles during pressing rather than on the reinforcing capacity of the fungal binder, which may explain the comparatively high water absorption (112.1%) and thickness swelling (126.33%) reported by Shakir *et al.* [30].

Genetic Regulation and Cell Wall Composition. The fungal cell wall is itself a nanocomposite structure consisting of crystalline chitin nanofibers embedded in a matrix of β-glucans and glycoproteins [29,37]. The ratio of crystalline chitin to amorphous glucan is a critical determinant of preform stiffness. Chen *et al.* [6] provided key insights into the genetic regulation of these components under different cultivation regimes. By comparing mycelium grown on solid lignocellulosic substrates (Mat-S) *versus* liquid nutrient cultures (Mat-L), they observed significant differential expression of genes related to cell wall synthesis. The liquid-cultured mycelium exhibited upregulation of chitin synthase genes, resulting in biomass with approximately 8% chitin content and an elastic modulus of 3416.6 ± 196.4 MPa after ultra-high-pressure pressing— significantly higher than the solid-state counterpart (10.3 MPa tensile strength). This genetic plasticity implies that the biological interface is a programmable variable: by manipulating culture conditions, it is possible to modulate the fungus’s chitin output, thereby engineering a stiffer, more thermally stable reinforcement phase prior to pressing.

Environmental Modulation. The incubation environment acts as an external signal that steers the fungal developmental program. Carbon dioxide (CO_2) concentration is a particularly potent regulator of

fungal morphology. In nature, elevated CO_2 indicates that the mycelium is deep within a substrate, triggering vegetative hyphal extension. Low CO_2 signals proximity to the surface, triggering the formation of reproductive structures (mushroom primordia). In composite manufacturing, this mechanism is exploited by maintaining elevated CO_2 concentrations (typically 5%–7%) during incubation, suppressing mushroom formation—which would constitute structural defects in a homogeneous board—and promoting uniform vegetative hyphal proliferation throughout the substrate [5]. However, the balance is critical: excessive CO_2 (> 15%) can inhibit aerobic respiration and stall growth entirely, creating zones of uncolonized substrate that become weak points upon pressing.

Temperature also plays a regulatory role beyond simple growth rate optimization. While constant temperatures of 25–30 ℃ are standard [5], Livne *et al.* [7] demonstrated that diurnal temperature cycling can enhance material properties. Periodic thermal stress may induce hyphal wall thickening or the production of stress-response proteins, creating a more resilient biological interface prior to the demanding conditions of heat pressing—though the specific mechanisms remain to be elucidated.

1.5.2. Nutritional and substrate conditions

Carbon-to-Nitrogen Ratio. The substrate serves a dual function as both structural aggregate and nutritional source. The Carbon-to-Nitrogen (C:N) ratio is a principal governor of fungal metabolism: a range of approximately 20:1 to 40:1 is generally considered optimal for the vigorous vegetative growth required for composite formation [1]. Carbon, derived principally from cellulose, hemicellulose, and lignin, provides energy for respiration and the carbon skeletons for cell wall polysaccharides, while nitrogen is the limiting factor for the synthesis of enzymes, proteins, and chitin.

Deviations from this range have structural implications. A high C:N ratio (nitrogen-poor substrate, e.g., pure sawdust) forces the fungus into a nutrient-scavenging mode; while this can stimulate the production of lignocellulolytic enzymes to access locked nutrients, the resulting hyphal network is often sparse and low in biomass, yielding a brittle preform with insufficient fungal material to bridge the gaps between substrate particles. Conversely, a low C:N ratio (nitrogen-rich substrate, e.g., supplemented with wheat bran or malt extract) promotes rapid, dense hyphal proliferation and increases the chitin and protein content available for thermal cross-linking. Yang and Qin [32] achieved their exceptional tensile strengths (12.99 MPa) using a substrate blended with coffee grounds—a nitrogen-rich additive—suggesting that the nutritional enhancement of the preform contributed to the final composite performance alongside their novel slurry preparation method.

However, nitrogen supplementation introduces a processing risk. A nitrogen-rich environment is highly conducive to contamination by opportunistic molds (Trichoderma, Aspergillus) and bacteria, which can outcompete the target basidiomycete and compromise the structural integrity of the biological interface. Nutritional optimization therefore represents a balancing act: providing sufficient nitrogen to maximize hyphal density and chitin yield without crossing the threshold that invites competitive organisms or suppresses the very enzymes needed for interface activation.

Enzymatic Pre-treatment and Interface Activation. The formation of the biological interface is not a passive deposition of hyphae onto inert surfaces but an active biochemical remodeling of the substrate. White-rot fungi secrete a suite of extracellular oxidative enzymes—laccase, lignin peroxidase (LiP), and manganese peroxidase (MnP)—that partially depolymerize lignin [8]. In the context of composite engineering, the goal is partial modification rather than complete degradation: the enzymes generate

radical species within the lignin polymer network, cleaving select bonds while leaving others intact, increasing the concentration of reactive functional groups—particularly phenolic hydroxyls—on the surface of substrate particles.

This enzymatic activation is functionally a biological pre-treatment that primes the interface for thermal bonding. Enzymatically modified lignin, having been partially depolymerized, exhibits a lower effective glass transition temperature and increased chemical reactivity compared to native lignin [11]. When the composite is subsequently subjected to heat pressing, this activated lignin softens and flows more readily, acting as a superior natural adhesive. The nutritional conditions directly regulate this enzymatic activity: high concentrations of easily accessible carbon (glucose) or nitrogen can repress the expression of ligninolytic genes through catabolite repression. To maximize interface activation, the substrate must be formulated to induce a degree of nutrient limitation at the appropriate growth phase, triggering the release of lignin-modifying enzymes before the material is harvested for pressing.

Additives and Trace Minerals. The enzymatic machinery of the interface can be further modulated through substrate additives. Calcium carbonate ($CaCO_3$) or gypsum can buffer the pH against the organic acids (primarily oxalic acid) produced during fungal metabolism, maintaining conditions in the optimal range (pH 5.0–7.0) for sustained growth; calcium ions also play a role in hyphal tip extension and branching, with their presence correlated with increased branching density and thus more extensive mechanical interlocking [33]. Additionally, ligninolytic enzymes require metal co-factors—laccase requires copper, while MnP requires manganese—and trace supplementation of these minerals can enhance enzymatic activity, leading to more effective surface activation of the lignocellulosic particles prior to pressing.

1.5.3. Particle size and scale effects on colonization

The Diffusion–Surface Area Trade-off. The physical geometry of the substrate—particle size, distribution, and inter-particle void volume—dictates the kinetics and completeness of colonization and therefore the spatial uniformity of the biological interface. A fundamental trade-off exists between surface area availability and oxygen diffusion. Fine particles (< 2 mm) offer a massive specific surface area for fungal attachment, leading to dense, cohesive hyphal networks. However, fine particles pack tightly, creating small interstitial voids with high tortuosity that severely restrict oxygen diffusion into the bulk material. Because mycelium is aerobic, if oxygen cannot penetrate to the core, the fungus colonizes only the exterior, leaving the interior unbound—a phenomenon that can be termed a “dead core” effect. Heat-pressed boards with uncolonized interiors will exhibit weak internal bonding and are prone to delamination under mechanical load.

Coarse particles (> 5 mm) create substrate beds with large interstitial voids and high porosity, facilitating gas exchange and enabling uniform colonization of thick sections. However, the specific surface area available for bonding is significantly lower, and large particles are more difficult for mycelium to fully encapsulate, potentially creating “islands” of unbound lignocellulose that act as stress concentrators during mechanical loading. Empirical studies suggest an optimal particle size window between approximately 2–5 mm for general composite applications [15,9]. Within this range, the void fraction is sufficient to support aerobic respiration throughout the matrix, yet the particles are small enough to be effectively bound by the hyphal network.

Notably, a graded particle size distribution—combining coarse and fine fractions—may be superior to a uniform particle size for heat-pressed boards. Coarse particles provide the skeletal structure and gas diffusion pathways during growth, while fine particles fill the interstices, maximizing the final density and minimizing residual voids during compression. This concept is well-established in conventional particleboard manufacturing but has not been systematically investigated for mycelium composites.

Void Fraction and Compressibility. The void space in the pre-pressed composite is a critical variable for the densification process. Mycelium composites are typically grown as low-density foams (170–270 kg/m^3) [1,27]. To achieve the densities of commercial fiberboards (> 800 kg/m^3), the material must undergo compression ratios often exceeding 3:1. The particle size distribution determines the initial packing density and the nature of the voids. If the mycelium grows too densely within large interstitial voids—forming a felted mass—it may trap air or moisture. During hot pressing, this trapped vapor can generate high internal steam pressures. If the matrix permeability is too low to allow steam egress, the board may suffer from blowouts or blisters upon press opening. The particle architecture must therefore be engineered to balance biological requirements (oxygen ingress during growth) with processing requirements (steam egress during pressing). The interaction between particle morphology—fibrous, flake-like, and granular forms—and pressing outcomes is discussed further in Section 3.9.1.

1.5.4. Implications for interfacial structure

The biological processes described above establish the precursor state of the interface prior to heat pressing. Before any thermal energy is applied, the composite is held together by two “cold” binding mechanisms: (1) mechanical interlocking, whereby branching hyphae physically penetrate the pores, lumens, and surface irregularities of substrate particles [5], and (2) biochemical adhesion, mediated by surface glycoproteins (adhesins) and hydrophobins that facilitate molecular adhesion between the fungal cell wall and the lignocellulosic substrate [3]. FTIR analyses of colonized substrates have confirmed hydrogen bonding between chitin and lignocellulosic components prior to pressing, evidenced by shifts in hydroxyl stretching bands (3200–3400 cm^{-1}) and amide peaks (1630–1660 cm^{-1}) [43,9].

These pre-existing chemical interactions are then consolidated and amplified by the thermochemical mechanisms detailed in Sections 1.4.1–1.4.3: lignin plasticization fills remaining voids and wets the hyphal surfaces, Maillard cross-linking creates covalent bonds between fungal proteins and plant polysaccharides, and the chitin reinforcement maintains its crystalline integrity throughout the process. Crucially, the strength of the pressed composite is not determined solely at the moment of pressing but is substantially pre-programmed during the colonization phase. A preform with high chitin content, dense hyphal penetration, and enzymatically activated substrate surfaces will respond to thermal processing with superior bonding compared to a sparsely colonized material with unmodified lignin.

The density and architecture of the hyphal network also governs how effectively pressure is transmitted through the material during compaction. A thoroughly colonized composite distributes compressive stress more uniformly, reducing the formation of density gradients and weak zones that serve as crack initiation sites. Conversely, composites with superficial colonization—where hyphae form a dense surface mat but fail to penetrate the interior—may develop a strong skin but a weak core, analogous to the density profiles observed in poorly pressed conventional particleboard.

However, the thermal processing that consolidates these biological bonds also introduces trade-offs. Heat pressing at temperatures above 160 ℃ denatures hydrophobin surface proteins [6], eliminating the

natural water repellency conferred by the biological phase and rendering the composite hydrophilic despite its higher density (see Section 3.9.2). This implies that optimizing the biological interface for maximum mechanical performance may require accepting compromises in other functional properties, or alternatively, engineering fungal strains with enhanced hydrophobin expression or thermal stability—an avenue of genetic optimization that remains unexplored.

The complex interplay between growth regulation, nutritional optimization, substrate geometry, and interfacial chemistry underscores that heat pressing performance cannot be understood in isolation from the biological processes that precede it. The pre-pressing biological preform represents a multi-dimensional design space—encompassing species selection, incubation regime, substrate formulation, and particle architecture—that remains largely unexplored in systematic fashion, particularly with respect to its interaction with pressing parameters. Future research that simultaneously optimizes biological and thermomechanical variables, rather than treating them sequentially, offers the greatest potential for advancing mycelium composite performance toward commercial viability.

2. Methods

This integrative review follows a structured approach to aggregate and analyze quantitative data from studies on heat pressing mycelium composites. The methodology is designed to ensure a comprehensive and systematic analysis of the available literature.

2.1. Search strategy

Due to the heterogeneity in terminology within this emerging field, a multi-faceted search strategy was employed to ensure comprehensive coverage while maintaining specificity. The Scopus database was selected as the primary source due to its extensive coverage of materials science and biotechnology literature, superior filtering capabilities, and more manageable result sets compared to broader academic search engines.

Searches were conducted using Publish or Perish software (version 8) in January 2026. Four complementary queries were designed to capture different aspects of heat-pressed mycelium composite research:

(1) Query 1 (Explicit thermal processing): mycelium AND ("heat press*" OR "hot press*" OR "thermal press*" OR "pressing temperature") NOT textile*

(2) Query 2 (Pressed composites): mycelium AND composite* AND press* NOT (textile* OR fabric* OR gene OR expression OR soil OR disease OR oil OR extract OR enzyme OR biocontrol OR bacteria)

(3) Query 3 (Structural applications): mycelium AND (board* OR panel* OR "building material*") AND (mechanical OR strength OR modulus)

(4) Query 4 (Comprehensive studies): mycelium AND ("biocomposite*" OR "bio-composite*" OR lignocellulosic) AND (properties OR factors OR processing OR review OR insight) NOT (textile* OR fabric* OR gene OR expression OR soil OR disease)

This multi-query approach was necessary because studies in this field use inconsistent terminology—some explicitly mention "heat pressing" or "hot pressing" in titles and keywords, while

others describe the same process as "thermal treatment", "pressing at elevated temperature" or simply "pressing" with temperature parameters detailed only in methods sections.

2.2. Selection criteria

Inclusion criteria:

- Publication years: 2018–2026
- Research focus: Production of heat pressed mycelium composites
- Language: English
- Data requirements: Quantitative data on at least one of the following properties after heat pressing: tensile strength, flexural strength, or elastic modulus

Exclusion criteria:

- Paper type: Reviews
- Research focus: Mycelium materials not classified as composites; mycelium composites without heat pressing; heat pressing of other materials
- Data type: Qualitative data only

The initial search yielded 421 references across all four queries. Results were exported and imported into Rayyan, a web-based systematic review management tool, for deduplication and screening. After deduplication, 130 duplicate entries were identified and removed, resulting in 291 unique references for screening, which were manually screened based on title and abstract, resulting in 18 studies for full assessment, out of which 14 were ultimately included in the review.

Figure 5 presents the PRISMA flow diagram detailing the literature search and study-selection process, from initial database query results through deduplication, title-and-abstract screening, and full-text assessment to the final set of included studies.

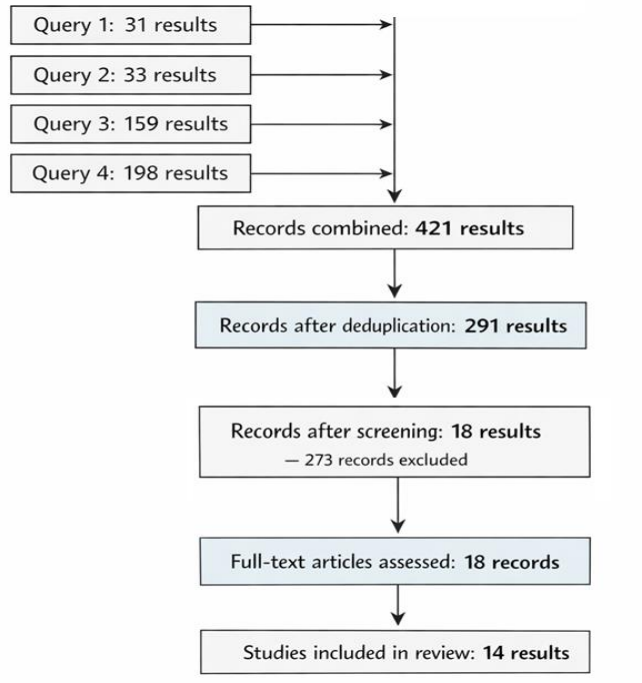

Figure 5. PRISMA diagram.

2.3. Data extraction and analysis

Given the heterogeneity inherent in this emerging field, a straightforward aggregation approach was adopted to maximize transparency and enable identification of broad trends across studies rather than precise effect estimation.

Quantitative data were manually extracted from each study and compiled into a standardized spreadsheet. For each experimental condition reported, we recorded the processing parameters—temperature (℃), pressure (MPa), and time (minutes)—alongside the mechanical properties measured: tensile strength (MPa), elastic modulus (MPa), flexural strength (MPa), density (kg/m^3), and internal bonding strength (MPa). Where studies reported pressure in kilonewtons rather than megapascals, values were converted using the sample dimensions provided. When studies reported multiple experimental conditions, all conditions with complete parameter-property pairs were included as separate observations to preserve the within-study variation that might reveal parameter-property relationships. The final dataset comprised 67 observations across 14 studies.

Data completeness varied substantially across properties, reflecting the lack of standardized reporting conventions in mycelium composite research. Processing parameters were reported for > 95% of observations. Among mechanical properties, tensile strength (n = 54), elastic modulus (n = 58), and density (n = 56) were well-represented, while flexural strength (n = 13) and internal bonding (n = 7) were sparsely reported. This uneven coverage is quantified in the co-reporting matrix (Section 3.3), which reveals that certain property pairs—such as tensile strength and internal bonding—were never measured together in any single study.

Statistical analysis was performed using Python with the Pandas library. Pearson correlation coefficients were calculated for all parameter-property pairs using the DataFrame.corr() method, which employs pairwise complete observations—that is, each correlation coefficient was computed using all observations with non-missing values for both variables in the pair, regardless of missing values in other columns. This approach maximizes data utilization given the fragmented reporting across studies; requiring complete cases across all variables would have reduced sample sizes to impractical levels.

Pearson correlation was selected as the primary measure for several reasons. First, visual inspection of scatter plots (not shown) revealed approximately linear relationships between processing parameters and mechanical properties within the ranges studied, without obvious curvilinear patterns that would favor rank-based methods. Second, Pearson coefficients are more interpretable for continuous engineering variables and align with how parameter-property relationships are typically reported in materials science literature. Third, given the exploratory nature of this analysis—where the goal was to identify general trends rather than establish precise predictive models—the choice between Pearson and Spearman correlations was unlikely to alter conclusions substantially. Indeed, supplementary analysis using Spearman rank correlations yielded similar patterns of significance (data not shown). Significance was assessed at $\alpha = 0.05$ using two-tailed tests.

Several important caveats apply to the interpretation of these correlations. The observational nature of aggregated cross-study data means that correlations reflect associations confounded by multiple uncontrolled variables—including fungal species, substrate composition, particle size distribution, moisture content, sample geometry, and testing protocols—that varied systematically across studies. For instance, the counterintuitive negative correlation between temperature and density (Section 3.2) likely

reflects confounding from the Yang and Qin [32] dataset, which achieved high mechanical properties at lower temperatures through a fundamentally different slurry-based preparation method. Similarly, the dominance of this single comprehensive study (45 of 67 observations) in the tensile strength, elastic modulus, and density measurements means that correlations for these properties are heavily weighted toward one laboratory's methods and materials.

Consequently, the correlation analysis should be interpreted as hypothesis-generating rather than hypothesis-confirming. The consistent significance of pressure across all mechanical properties suggests densification as a primary driver of performance—a finding supported by theoretical understanding of composite mechanics—but definitive causal claims would require controlled experiments that systematically vary single parameters while holding others constant. The substantial heterogeneity quantified throughout the Results section (coefficient of variation > 45% for most properties; Section 3.1) underscores that between-study variation exceeds what can be explained by processing parameters alone, highlighting the urgent need for standardized protocols discussed in Section 3.6.

Visualization was performed using Seaborn and Matplotlib. The correlation heatmap (Figure 6) displays Pearson coefficients for all variable pairs, while scatter plots of parameter space coverage (Figure 8) and property heatmaps (Figure 9) were generated to contextualize the numerical findings.

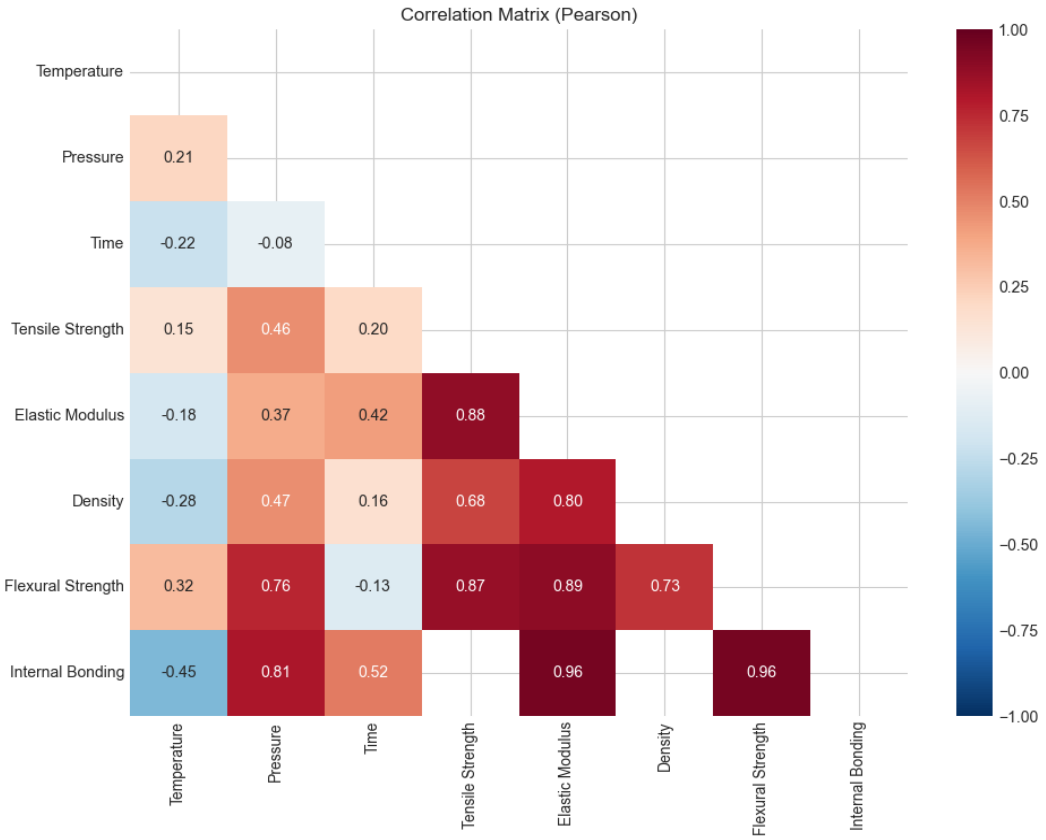

Figure 6. Correlation matrix of parameters and properties.

3. Results

A summary of the selected studies is present in Table 1. It includes the species and substrate used, pressing parameters, and measurements taken. When multiple values were reported, at least two datapoints were selected for comparison, including the one with maximal values.

Table 1. Results and parameters of selected studies.

Study	Title	Species and Substrate	Pressing	Results
Appels, *et al.*, 2019	Fabrication factors influencing mechanical, moisture- and water-related properties of mycelium-based composites	*P. ostreatus*, *T. multicolor* Rapeseed straw, Beech sawdust, Coton	150 ℃ 20 min 30 kN (on 34 × 34 cm, equivalent to 0.26 MPa)	Unpressed (foam-like) • Tensile strength—0.05 MPa • Elastic modulus—13 MPa • Flexural Strength—0.24 MPa • Density—170 kg/m ^{3} Hot pressed • Tensile strength—0.24 MPa • Elastic modulus—97 MPa • Flexural Strength—0.87 MPa • Density—390 kg/m ^{3}
Nussbaumer, *et al.*, 2023	Material characterization of pressed and unpressed wood-mycelium composites derived from two *Trametes* species	*T. pubescens*, *T. versicolor* Beech sawdust	120 ℃ 50 min 100 kN (on 8 cm diameter disks, equivalent to 19.89 MPa)	Unpressed (foam-like) • Tensile strength—0.05 MPa • Elastic modulus < 1 MPa • Density—270 kg/m ^{3} Hot pressed • Tensile strength—0.35 MPa • Elastic modulus—14 MPa • Density—515 kg/m ^{3}
Livne, *et al.*, 2024	Increased CO_2 fixation and reduced embodied energy of mycelium bio-composite materials grown on a mixed substrate over diurnal temperature cycles	*T. hirsuta* Wheat straw and cellulose	150 ℃ 5 min 28.25 MPa	• Elastic modulus—50 MPa • Compression strength (at 20% strain)—5.6 MPa • Density—527 kg/m ^{3}
Liu, *et al.*, 2019	Preparation of a kind of novel sustainable mycelium/cotton stalk composites and effects of pressing temperature on the properties	*Ganoderma lucidum* Cotton stalk	160–200 ℃ 3 min 3.5–4.0 MPa	200 ℃ • Flexural strength—4.6 MPa • Elastic modulus—680 MPa • Internal bond—0.18 MPa • Density—600 kg/m ^{3} 160 ℃ • Flexural strength—1.4 MPa • Elastic modulus—260 MPa • Internal bond—0.05 MPa • Density—600 kg/m ^{3}
Liu, *et al.*, 2020	Improvement of mechanical properties of mycelium/cotton stalk composites by water immersion	*G. lucidum* Cotton stalk	200 ℃ 6 min 3.5–4.0 MPa	Dry hot pressing • Flexural strength—4.4 MPa • Elastic modulus—690 MPa • Density—600 kg/m ^{3} Hot pressing at 30% hydration • Flexural strength—6 MPa • Elastic modulus—860 MPa • Internal bond—0.34 MPa • Density—600 kg/m ^{3}
Elsacker, *et al.*, 2021	Mechanical characteristics of bacterial cellulose-reinforced mycelium composite materials	*T. versicolor* Bacterial Cellulose and Hemp	200/70 ℃ 10 h 30 kN (on 18.5 × 18.5 cm, ~0.87 MPa)	200 ℃ • Tensile strength—0.056 MPa • Flexural strength—2.94 MPa • Elastic modulus—7 MPa • Density—456 kg/m ^{3} • 70 ℃ • Tensile strength—0.034 MPa • Flexural strength—1.91 MPa • Elastic modulus—5 MPa • Density—531 kg/m ^{3}
Sun, *et al.*, 2022	Insight into mycelium-lignocellulosic biocomposites—essential factors and properties	*T. versicolor* Yellow birch	180 ℃ 8 min No pressure control	• Flexural strength—5.0 MPa • Elastic modulus—900 MPa • Density—640 kg/m ^{3}
Shakir, *et al.*, 2020	Preparation and Characterization of Mycelium as a Bio-Matrix in Fabrication of Bio-Composite	*P. ostreatus* Rubberwood Sawdust	130 ℃ 20/40 min 5 MPa	40 min • Flexural strength—3.91 MPa 20 min • Flexural strength—1.82 Mpa

Table 1. *Cont.*

Study	Title	Species and Substrate	Pressing	Results
Yang and Qin, 2023	Mycelium-based wood composites for light weight and high strength by experiment and machine learning	*P. eryngii* Hardwood sawdust with coffee grounds	80, 90, 100 ℃ 1, 2, 4, 8, and 16 h 6.75, 13.51, and 20.27 MPa	80 ℃, 1 h, 6.75 MPa • Tensile strength—5.64 MPa • Elastic modulus—1477 MPa • Density—1028 kg/m ^{3} 100 ℃, 4 h, 20.27 MPa • Tensile strength—12.99 MPa • Elastic modulus—3664 MPa • Density—1258 kg/m ^{3}
Chen *et al.*, 2025	Structural, Mechanical, and Genetic Insights into Heat-Pressed Fomes fomentarius Mycelium	*F. fomentarius* Hemp shives (Mat-S) Liquid culture (Mat-L)	160 ℃ 5 min 100 MPa	Mat-L: • Tensile strength 12.5 ±6.1 MPa, • Elastic modulus 3416.6 ±196.4 MPa; Mat-S: • Tensile strength 10.3 Mpa
Shakir *et al.*, 2025	Development and characterization of fiber-reinforced biopolymer composites from rice husk waste with Rhizopus oligosporus as a bio-binder	*R. oligosporus* Rice husk	130 ℃ 30 min 5 MPa	• Flexural strength—5.79 MPa • Tensile strength—2.59 MPa
Yan *et al.*, 2024	Formaldehyde-Free Bio-composites Based on Pleurotus ostreatus Substrate and Corn Straw Waste	*P. ostreatus* Spent substrate grown on corn stalk	190 ℃ 2 h 18 MPa	• Internal bonding strength—2.16 MPa
Khoo *et al.*, 2020	Development of formaldehyde-free bio-board produced from mushroom mycelium and substrate waste	*G. lucidum*, *P. ostreatus*, *A. polytricha* Spent mushroom substrates grown on rubber tree sawdust	160 ℃ 20 min 10 MPa	*G. lucidum* • Internal bonding strength—2.5 MPa *P. ostreatus* • Internal bonding strength—2.25 Mpa *A. polytricha* • Internal bonding strength—1.6 Mpa
Song *et al.*, 2025	Optimization of semi-wet hot-pressing conditions for Salix psammophila-based mycelium-bound boards via response surface methodology	Unspecified white-rot fungi *Salix psammophila*	194 ℃ 31 min 10 MPa Semi-wet process (30%–50% moisture content)	• Flexural strength—37.64 MPa

3.1. Statistical analysis

Table 2 presents summary statistics for the conventionally heat-pressed samples. Mean tensile strength was 6.312 MPa (SD = 3.074), with values ranging from 0.034 to 12.985 MPa. Elastic modulus averaged 2137.5 MPa (SD = 1082.5), spanning from 1.0 to 3664.8 MPa. Density ranged from 0.170 to 1.542 g/cm ^{3} with a mean of 1.182 g/cm ^{3}. Flexural strength showed considerable variability (mean = 5.886 MPa, SD = 9.726), reflecting both the sparse reporting of this property and the inclusion of the exceptional 37.64 MPa value from Song *et al.* [18]. Internal bonding strength, though based on limited data points, averaged 1.297 MPa (SD = 1.073).

What stands out most prominently is the substantial disparity in mechanical properties across studies, even among those employing ostensibly similar methodologies. For instance, tensile strength measurements varied from 0.034 MPa [37] to 0.35 MPa [35] among conventional pressing studies—a ten-fold difference. Similar discrepancies were observed for elastic modulus: Elsacker *et al.* [37] reported 5–7 MPa, Nussbaumer *et al.* achieved 14 MPa, while Liu *et al.* [15] obtained 860 MPa under optimized moisture conditions. The coefficient of variation for most properties exceeds 45%, indicating substantial heterogeneity that cannot be attributed solely to differences in processing parameters.

These variations likely stem from multiple confounding factors: differences in fungal species and their chitin content, substrate composition and particle size, moisture content during pressing, sample preparation for mechanical testing, and the testing protocols themselves. The lack of standardized testing procedures—comparable to ASTM or ISO standards used for conventional composites—makes direct comparisons between studies problematic and highlights an urgent need for methodological standardization in mycelium composite research.

Table 2. Mean values of mechanical characteristics.

	Tensile Strength[a]	Elastic Modulus[a]	Density[b]	Flexural Strength[a]	Internal Bonding[a]
Mean	6.312	2137.529	1.182	5.886	1.297
SD	3.074	1082.499	0.383	9.726	1.073
Min	0.034	1.000	0.170	0.240	0.050
Max	12.985	3664.800	1.542	37.640	2.500

[a] MPa; [b] g/cm ^{3}

As for a direct comparison between before and after hot pressing (Table 3), notably, only two studies [1,27] reported paired comparisons of unpressed and pressed materials from identical substrate/species combinations, limiting the ability to precisely quantify the effects of heat pressing independent of other experimental variables. Therefore, the data is limited by small sample sizes (N = 1–2), but it demonstrates significant improvements in mechanical properties through heat pressing. The most significant enhancement is seen in elastic modulus, showing a mean improvement of 973%. Tensile strength also showed substantial improvement, averaging 490%. Despite the limited dataset, the positive improvements are consistent across all properties.

Table 3. Improvements obtained by heat pressing.

	Tensile Strength	Elastic Modulus	Density	Flexural Strength
Mean Improvement	490%	973%	110%	263%
Min Improvement	380%	646%	91%	
Max Improvement	600%	1300%	129%	
N	2	2	2	1

3.2. Correlation analysis

Pearson correlation analysis revealed distinct relationships between processing parameters and mechanical properties (Table 4). Pressure emerged as the strongest and most consistent predictor, showing significant positive correlations with all five mechanical properties: tensile strength ($r = 0.463$, $p < 0.001$), elastic modulus ($r = 0.373$, $p < 0.01$), density ($r = 0.467$, $p < 0.001$), flexural strength ($r = 0.763$, $p < 0.01$), and internal bonding ($r = 0.813$, $p < 0.05$).

Temperature showed weaker and less consistent relationships. Only the negative correlation with density reached significance ($r = -0.283$, $p < 0.05$), while correlations with other properties were not statistically significant. This counterintuitive finding likely reflects confounding from the Yang and Qin [32]

dataset, which achieved high mechanical properties at lower temperatures (80–100 ℃) through extended processing and slurry preparation.

Table 4. Correlation statistics between processing parameters and mechanical properties.

Parameter	Property	n	r	p-value	Significance
Temperature	Tensile Strength	54	0,148	0.2854	
Temperature	Elastic Modulus	58	−0,176	0.1872	
Temperature	**Density**	**56**	**−0,283**	**0.0343**	*
Temperature	Flexural Strength	13	0,319	0.2875	
Temperature	Internal Bonding	7	−0,446	0.3159	
Pressure	**Tensile Strength**	**54**	**0,463**	**0.0004**	***
Pressure	**Elastic Modulus**	**57**	**0,373**	**0.0042**	**
Pressure	**Density**	**55**	**0,467**	**0.0003**	***
Pressure	**Flexural Strength**	**12**	**0,763**	**0.0039**	**
Pressure	**Internal Bonding**	**7**	**0,813**	**0.0263**	*
Time	Tensile Strength	54	0,199	0.1484	
Time	**Elastic Modulus**	**58**	**0,421**	**0.0010**	**
Time	Density	56	0,158	0.2446	
Time	Flexural Strength	13	−0,134	0.6614	
Time	Internal Bonding	7	0,522	0.2294	

Processing time demonstrated a significant positive correlation only with elastic modulus ($r = 0.421$, $p < 0.01$). Correlations with other properties did not reach significance, supporting observations that prolonged pressing does not necessarily improve outcomes.

The limited sample sizes for flexural strength (n = 12–13) and internal bonding (n = 7) reduce statistical power for these properties, and results should be interpreted cautiously. Nevertheless, the consistent significance of pressure across all properties suggests densification is a primary driver of mechanical performance in heat-pressed mycelium composites.

3.3. Data completeness and co-reporting

The heterogeneity of reporting across studies presented a significant analytical challenge. A co-reporting matrix (Figure 7) was constructed to quantify the overlap between measured properties across the dataset. Tensile strength, elastic modulus, and density demonstrated strong co-reporting, with 51–58 samples reporting pairs of these properties simultaneously. However, flexural strength was reported alongside tensile strength in only 5 samples, and alongside elastic modulus in only 9 samples. Internal bonding was the most sparsely reported property, with zero samples co-reporting both internal bonding and tensile strength, and only 3 samples reporting internal bonding alongside elastic modulus or flexural strength.

This fragmented reporting pattern limits the ability to establish comprehensive structure-property relationships and underscores the need for standardized testing protocols in mycelium composite research. The strong co-reporting among tensile strength, elastic modulus, and density reflects the dominance of the Yang and Qin [32] dataset, which systematically measured these three properties across 45 experimental conditions.

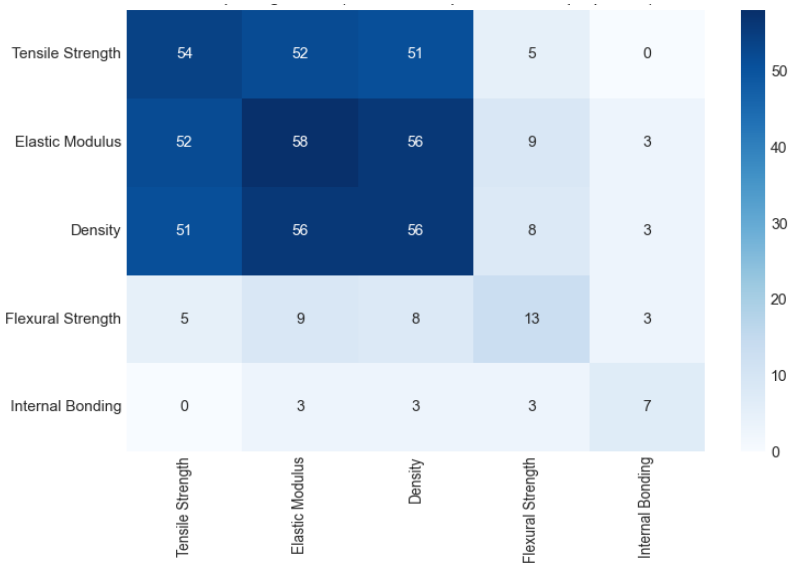

Figure 7. Co-reporting matrix.

3.4. Parameter space coverage and research gaps

Visualization of the experimental parameter space (Figure 8) reveals substantial clustering. The majority of studies operated within a narrow band of pressures (5–20 MPa), with processing times ranging from minutes to hours. The dense cluster at 80–100 ℃ and 6.75–20.27 MPa represents the systematic Yang and Qin [32] study.

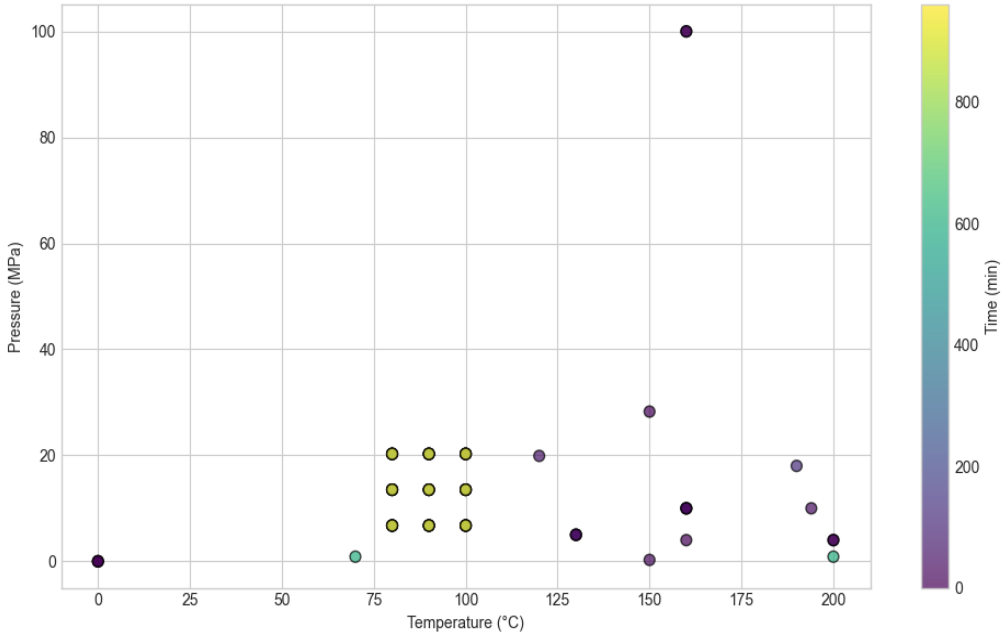

Figure 8. Parameter space coverage.

A single outlier at 160 ℃ and 100 MPa [6] represents recent exploration of ultra-high-pressure processing, achieving tensile strengths of 12.5 MPa—comparable to the slurry-based approach of Yang and Qin but through fundamentally different mechanisms.

Notably absent from the literature are systematic investigations of: (1) high temperature (> 170 ℃) combined with high pressure (> 20 MPa) processing, (2) the intermediate temperature range of 120–150 ℃

with pressures exceeding 20 MPa, and (3) short processing times (< 10 minutes) at elevated pressures. These gaps represent opportunities for future optimization studies, particularly given the industrial need for rapid processing cycles.

3.5. Discussion of key studies

Several studies stand out for their methodological innovations that suggest promising directions for future research.

3.5.1. Moisture optimization

The experimental work of Liu *et al.* [15] provides crucial empirical evidence for the moisture-dependent thermochemical mechanisms previously detailed in Section 1.4.1. Their findings confirm that a hydration level of 30% acts as a "sweet spot" for mechanical performance, directly leveraging the plasticization effect to lower the lignin glass transition temperature and facilitate the wetting of the chitinous reinforcement.

The quantitative impact of this optimization is substantial: at 30% hydration, Liu *et al.* achieved a flexural strength of 6 MPa and an elastic modulus of 860 MPa, compared to 4.4 MPa and 690 MPa in dry conditions. Beyond the chemical bonding advantages, the researchers highlighted a "steam shock" effect, where internal moisture facilitates rapid heat transfer into the core of the composite, ensuring superior compaction. These results, combined with their earlier optimization of temperature (200 ℃) and pressure (3.5–4.0 MPa), establish a practical framework for the "semi-wet" processing now being adopted in the field.

3.5.2. Substrate blending

The work by Yang and Qin [32] represents a significant departure from conventional processing methods. Rather than pressing intact mycelium-colonized substrate, they blended the material into a slurry before pressing. This approach yielded outlying results—tensile strength of 12.99 MPa and elastic modulus of 3664 MPa at 100 ℃ and 20.27 MPa pressure. Notably, even their control samples of pure vegetable matter achieved 8.8 MPa tensile strength, suggesting that the mechanical disruption of the substrate structure may be as important as the mycelial binding.

Analysis of their systematic parameter variations (Figure 9) reveals distinct patterns for each property. For elastic modulus, pressure exerted a clear dominant effect: mean values increased progressively from approximately 2400–2500 MPa at 6.75 MPa to 2600–3200 MPa at 20.27 MPa across all temperatures, with the maximum (3243 MPa) achieved at 100 ℃ and 20.27 MPa. Temperature effects on elastic modulus were comparatively modest and non-monotonic.

Tensile strength showed a less consistent pattern, with peak values occurring at 90 ℃/13.51 MPa (8.13 MPa) and 100 ℃/20.27 MPa (8.62 MPa), suggesting potential interaction effects between temperature and pressure. Density followed expected trends, increasing with both temperature and pressure, though the 90 ℃ conditions produced slightly higher densities than 100 ℃ at equivalent pressures.

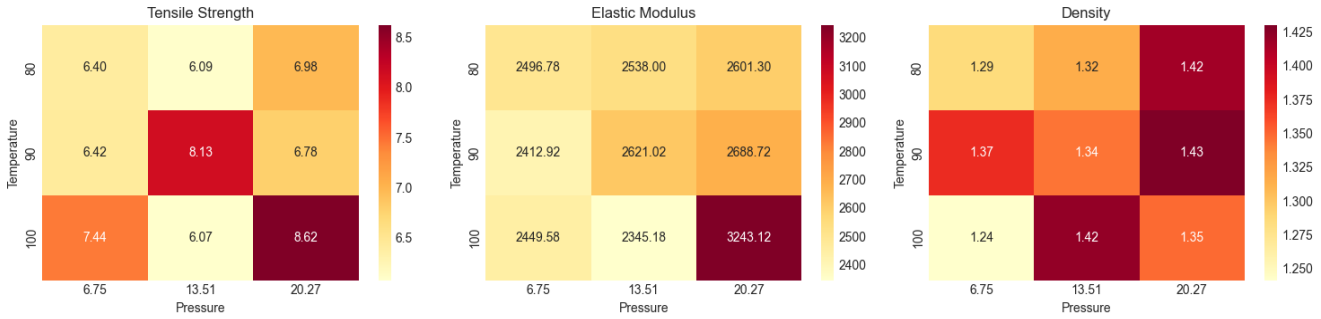

Figure 9. Yang & Qin heatmaps.

It is important to note that Yang and Qin's process operates at temperatures below the 130–170 ℃ range where chitin-lignin crosslinking is theorized to occur in conventional heat pressing. This suggests their material's strength derives from different mechanical and chemical mechanisms—the process more closely resembles paper-making or hardboard production, where fiber-fiber hydrogen bonding dominates rather than thermally-activated crosslinking. Their success with lower temperatures (80–100 ℃) but longer processing times (1–16 hours) also contrasts with the negative correlation between processing time and mechanical properties observed in higher-temperature studies. However, pressing times of up to 16 hours are largely unfeasible for industrial-scale production.

3.5.3. Bacterial cellulose reinforcement

Elsacker *et al.* [37] explored another innovative approach by incorporating bacterial cellulose, achieving modest improvements in mechanical properties while maintaining lower processing temperatures. Their work with split temperature profiles (200 ℃/70 ℃) offers insights into the potential for more nuanced thermal processing strategies. Bacterial cellulose nanofibers have a high surface area and crystallinity, providing additional sites for hydrogen bonding and acting as a scaffold that bridges gaps between larger substrate particles. This suggests that hybridizing mycelium with other biopolymers is a viable route to high-performance composites.

3.5.4. Ultra-high-pressure processing

Chen *et al.* [6] represents a pivotal advancement in mycelium composite research, utilizing ultra-high pressures (100 MPa) to create materials that rival ceramics and high-performance plastics. The researchers compared two distinct forms of *Fomes fomentarius* mycelium: Mat-S (solid state, traditional growth on lignocellulosic hemp shives) and Mat-L (pure fungal biomass harvested from liquid fermentation).

The Mat-L samples achieved a tensile strength of 12.5 ± 6.1 MPa and an elastic modulus of 3416.6 ± 196.4 MPa. This stiffness is comparable to bone or certain hard plastics and is significantly higher than the wood-composite analogues produced in earlier studies. Notably, the Mat-S (composite) samples were weaker (10.3 MPa), indicating that the lignocellulosic substrate, usually thought of as the reinforcement, may actually introduce defects or weak interfaces compared to the pure, dense hyphal network.

The study performed RNA sequencing and chemical analysis, revealing that the liquid-cultured mycelium had a higher chitin content (~8%) and lower β-glucan content (~18%) compared to the solid-state mycelium. The authors posit that the higher chitin concentration provides a stiffer crystalline backbone, which, when fused under 100 MPa, creates a superior material.

A critical finding for application is the loss of hydrophobicity. Unpressed mycelium is naturally water-repellent (contact angle > 130 °) due to surface proteins called hydrophobins. Heat pressing at 160 ℃ denatured these proteins, reducing the contact angle to < 30 ° for Mat-L. This implies that while heat pressing solves the mechanical strength issue, it exacerbates the water absorption issue, necessitating secondary coatings or chemical treatments.

This study suggests a bifurcation in the industry: (1) Composite Route using agricultural waste for bulk, low-cost panels (Strength ≈ 5 MPa); and (2) Pure Mycelium Route using liquid fermentation to produce pure mycelium "leathers" or "ceramics" for high-value applications (Strength > 12 MPa).

3.5.5. Spent mushroom substrate utilization

A parallel line of research has focused on utilizing spent mushroom substrate (SMS) as the primary feedstock for bio-board production, representing a circular economy approach that valorizes waste from the mushroom cultivation industry. Khoo *et al.* [4] demonstrated that SMS from various mushroom species could be converted into high-performance bio-boards without synthetic adhesives. Their work compared SMS from *Ganoderma lucidum*, *Pleurotus ostreatus*, and *Auricularia polytricha*, finding that *G. lucidum* SMS produced the highest internal bonding strength (2.5 MPa) when pressed at 160 ℃ and 10 MPa for 20 minutes. The superior performance of *G. lucidum* was attributed to its dense mycelial network and higher chitin content, which enhanced fiber-to-fiber bonding during heat pressing.

Yan *et al.* [31] extended this approach by combining *P. ostreatus* spent substrate with corn stalk waste, achieving internal bonding strengths of 2.16 MPa and notably low thickness swelling of 18.3% using a dry process at 190 ℃ and 18 MPa. Their work is particularly significant for demonstrating that the mycelium-coated substrate acts as a natural hydrophobic adhesive, addressing the inherent water sensitivity of lignocellulosic materials. The use of a dry process also offers environmental advantages over wet processing methods that require extensive water treatment.

3.5.6. Novel species exploration

While most mycelium composite research has focused on basidiomycete species such as *Pleurotus*, *Trametes*, and *Ganoderma*, Shakir *et al.* [30] investigated *Rhizopus oligosporus*—a zygomycete traditionally used in tempeh fermentation—as a bio-binder for rice husk composites. This species offers distinct advantages including rapid growth rates (optimal inoculation of only 4 days) and well-established industrial cultivation protocols.

Their composites achieved tensile strength of 2.59 MPa and flexural strength of 5.79 MPa when pressed at 130 ℃ and 5 MPa for 30 minutes. While the lower processing temperature (130 ℃ *versus* 160–200 ℃ in other studies) has implications for energy efficiency in industrial-scale production, the pressing time was longer than in many other papers.

However, the composites exhibited relatively high water absorption (112.1%) and thickness swelling (126.33%), indicating that the hydrophobic properties associated with basidiomycete hydrophobins may not be present in *R. oligosporus* mycelium. This trade-off between mechanical performance and water resistance highlights the need for species-specific optimization of post-processing treatments or protective coatings.

3.5.7. Process optimization via response surface methodology

Song *et al.* [18] represents a methodological advancement in mycelium composite research through the application of Box-Behnken response surface design to optimize hot-pressing parameters. Using *Salix psammophila* (sandy willow) as the substrate and white-rot fungal mycelium as a bio-adhesive, they systematically investigated the effects of pressure (6–10 MPa), temperature (170–200 ℃), and time (20–40 min) on board performance.

The study employed a semi-wet hot-pressing process with substrate moisture content controlled between 30–50%, building upon the moisture optimization principles identified by Liu *et al.* [15]. Under optimal conditions (10 MPa, 194 ℃, 31 min), the mycelium-bound boards achieved a modulus of rupture (MOR) of 37.64 MPa—a value that dramatically exceeds all other studies reviewed herein. For comparison, this is approximately 6 times higher than the best flexural strength achieved by Liu *et al.* [15] at 6 MPa, and approaches the mechanical performance of commercial medium-density fiberboard (MDF), which typically exhibits MOR values of 20–40 MPa.

The exceptional performance can be attributed to several factors: (1) the higher processing temperature (194 ℃) compared to most studies, which maximizes lignin flow and Maillard cross-linking; (2) the elevated pressure (10 MPa), which achieves greater densification; (3) the semi-wet process, which optimizes lignin plasticization while reducing energy consumption compared to wet processing; and (4) the use of *Salix psammophila*, a substrate with favorable fiber characteristics for board production. Importantly, the boards also demonstrated acceptable water resistance (48.1% water absorption, 23.01% thickness swelling), suggesting that the high-temperature processing did not completely compromise hydrophobic properties.

The response surface methodology employed by Song *et al.* provides a template for future optimization studies, enabling researchers to identify optimal processing windows while accounting for parameter interactions. Their finding that pressure and temperature have the most pronounced effects on performance aligns with the correlation analysis presented in this review (Section 3.2), providing independent validation of these relationships.

3.5.8. Synthesis of processing approaches

The studies reviewed herein reveal that mycelium composite performance is governed by a complex interplay of processing parameters, with no single “optimal” approach emerging. Instead, several distinct strategies have proven successful, each with particular advantages and trade-offs.

(1) Temperature-Pressure Relationships

A clear pattern emerges across studies: higher temperatures and pressures generally yield superior mechanical properties, but within bounded ranges. The conventional processing window of 130–170 ℃, established in earlier work, appears conservative in light of recent findings. Song *et al.* [18] demonstrated that temperatures approaching 200 ℃ (194 ℃ optimal) combined with moderate pressure (10 MPa) can achieve flexural strengths of 37.64 MPa—performance comparable to commercial MDF. This suggests the upper temperature limit may be extended when moisture content is properly controlled, as the steam generated facilitates heat transfer while the water plasticizes lignin at lower effective temperatures.

(2) The Critical Role of Moisture

Liu *et al.* [15] first identified 30% hydration as optimal for mechanical performance, a finding corroborated and extended by Song *et al.* [18], who formalized the "semi-wet" approach (30%–50% moisture). This moisture range appears to represent a thermodynamic sweet spot: sufficient water to plasticize lignin and facilitate hydrogen bonding, yet not so much as to require energy-intensive drying or cause steam-related defects. Notably, none of the high-performing studies employed truly dry pressing, suggesting that some moisture content is essential for achieving strong interfacial bonding.

(3) Ultra-High Pressure as an Alternative Pathway

Chen *et al.* [6] demonstrated that extreme pressures (100 MPa) can achieve high mechanical performance (tensile strength 12.5 MPa) at moderate temperatures (160 ℃), representing an alternative to high-temperature processing. However, this approach requires specialized equipment and sacrifices the material's natural hydrophobicity. The choice between high-temperature/moderate-pressure and moderate-temperature/ultra-high-pressure processing may ultimately depend on available equipment, energy costs, and end-use requirements for water resistance.

(4) Substrate and Species Effects

Performance varies substantially with substrate selection. Yang and Qin [32] achieved tensile strengths of 12.99 MPa using blended hardwood sawdust, while Song *et al.* [18] reached 37.64 MPa flexural strength with Salix psammophila. Shakir *et al.* [30] demonstrated that novel fungal species (Rhizopus oligosporus) can outperform traditional basidiomycetes at equivalent processing conditions, achieving 2.59 MPa tensile and 5.79 MPa flexural strength. These findings suggest that substrate-species optimization may offer performance gains comparable to processing parameter optimization.

(5) Process Optimization Methodologies

Two studies employed systematic optimization approaches rather than single-factor experiments. Yang and Qin [32] used machine learning to model the relationships between processing parameters and mechanical properties, while Song *et al.* [18] applied response surface methodology (Box-Behnken design). Both approaches successfully identified non-obvious parameter combinations and revealed significant parameter interactions. The convergence of their findings—that pressure and temperature are the dominant factors, with time playing a secondary role—provides robust validation of these relationships. These methodologies offer a path forward for industrial process development, enabling efficient optimization across the multi-dimensional parameter space.

(6) Practical Implications

For industrial applications, the following guidelines emerge from the aggregated data:

- Temperature: 160–200 ℃, with higher temperatures viable when moisture is controlled
- Pressure: 5–20 MPa for conventional processing; higher pressures yield diminishing returns without specialized equipment
- Time: 5–30 minutes; longer times show diminishing returns at higher temperatures
- Moisture: 30%–50% pre-pressing moisture content appears optimal
- Substrate: Fiber characteristics significantly impact final properties; woody substrates with favorable lignin content perform best

The range of successful approaches—from Chen *et al*'s ultra-high pressure processing to Song *et al.*'s optimized semi-wet method—demonstrates that multiple pathways exist for producing high-performance mycelium composites. The challenge now lies in selecting approaches that balance performance requirements with practical constraints of energy consumption, equipment availability, and production throughput.

3.6. Standardization of testing protocols

One of the most significant barriers to commercialization is the lack of standardized testing. Currently, researchers use arbitrary sample sizes (e.g., 8 cm disks *vs.* 34 cm squares) and varying crosshead speeds, rendering cross-study comparisons difficult. To legitimize mycelium composites as construction materials, the field must adopt and adapt established standards from the wood and plastics industries.

ASTM D1037 ("Standard Test Methods for Evaluating Properties of Wood-Base Fiber and Particle Panel Materials") is the most relevant standard for mechanical testing. For flexural strength (Modulus of Rupture), a three-point bending test with standard width 50 mm and span 150 mm (or 24 × thickness) should be employed. For internal bond (tensile strength perpendicular to surface), bonding metal blocks to the faces of the board and pulling them apart provides the ultimate test of the lignin-mycelium adhesion. Several recent studies have adopted internal bonding strength (IBS) as a primary metric, following wood-based panel standards. Khoo *et al.* [4] and Yan *et al.* [31] both reported IBS values that demonstrate the viability of mycelium composites for structural applications. The adoption of IBS testing alongside flexural and tensile measurements would provide a more complete characterization of mycelium composite performance and facilitate direct comparison with conventional wood-based panels.

For moisture resistance, ASTM D1037 Water Absorption and Thickness Swelling (24-hour soak) should be standard. While water absorption by weight is useful, thickness swelling is the critical metric for construction materials—a swelling of > 15% is typically a failure for particleboard.

To substantiate environmental claims, ISO 16929 ("Determination of the degree of disintegration of plastic materials under defined composting conditions") should be adopted, requiring > 90% disintegration within 12 weeks.

3.7. Life cycle considerations

While mycelium composites are frequently promoted for their sustainability advantages, comprehensive life cycle assessments (LCA) specific to heat-pressed variants remain limited. Recent studies on non-pressed or minimally processed mycelium composites provide important context. Fernandes *et al.* [40] reported that mycelium-based composites are associated with 0.3668 kg CO_2e/kg material in laboratory-scale production, demonstrating favorable climate change impacts compared to extruded polystyrene, foam concrete, and rockwool insulation. However, the same study noted that electricity consumption during incubation and drying contributes significantly to the overall environmental footprint, while land use and water demand may exceed those of conventional materials.

Wendt *et al.* [41] conducted a cradle-to-grave LCA of mycelium-based acoustic insulation panels and confirmed that energy use during the manufacturing stage—particularly incubation and drying—is the dominant contributor to global warming potential across most impact categories. A recent comprehensive review by Wang *et al.* [42] noted that mycelium-based composites can achieve net-negative global warming potential by sequestering more carbon during cultivation than is emitted throughout their life cycle, though energy-intensive processing steps pose significant hurdles to broader adoption.

The additional energy requirements of heat pressing—temperatures of 130–200 ℃, pressures of 5–100 MPa, and processing times ranging from minutes to hours—introduce further environmental burden beyond the baseline growth and drying phases.

Nevertheless, comparative analyses suggest that heat-pressed mycelium composites may still offer substantial environmental benefits over conventional alternatives. Critically, traditional particleboard and MDF production also requires heat pressing at comparable temperatures (typically 140–220 ℃) to cure the synthetic adhesives, meaning the thermal processing energy is not unique to mycelium composites—but the adhesive production is. A recent study by Ding *et al.* [43] directly compared formaldehyde-free mycelium-based biocomposites with conventional particleboard production and found that replacing synthetic adhesives with mycelium-based binding decreased global warming potential by 81.22%. This dramatic reduction reflects the elimination of petroleum-derived resin synthesis, which carries substantial upstream emissions from petrochemical feedstock extraction and processing.

Future research should systematically compare the cradle-to-grave environmental footprints of heat-pressed mycelium boards against both unpressed variants and conventional particleboards under standardized production conditions. Such assessments should account for the full process chain, including substrate sourcing, sterilization, incubation energy, pressing energy, and end-of-life scenarios, to validate sustainability claims for industrial-scale applications.

3.8. Limitations

This systematic review faced methodological limitations that should be considered when interpreting its findings. The primary one was the insufficient number of studies providing direct comparisons between pressed and unpressed mycelium composites. While the original aim was to quantify the specific effects of heat pressing, the scarcity of papers reporting pre- and post-pressing measurements prevented a comprehensive comparative analysis. Although excluding studies without such comparative data would have improved methodological rigor, it would have resulted in an impractically small sample size. Therefore, these studies were retained to provide broader insights, albeit at the cost of sample homogeneity.

A second significant limitation is that studies often exhibited considerable variation in their experimental approaches, creating a methodological heterogeneity that made direct comparisons challenging. Differences in fungal species selection, substrate composition, processing parameters, and testing protocols contributed to high variability in reported outcomes. While this heterogeneity complicated efforts to draw definitive conclusions about optimal processing conditions or expected performance improvements, the review does hint at possible directions for subsequent research.

Moreover, there are potential sources of bias that may affect the findings of this review. Publication bias likely favors studies showing positive results from heat pressing, potentially overestimating its benefits. Additionally, researcher bias may be present in studies funded by companies developing mycelium materials, though other than utilizing material from such companies, none of the included studies declared such conflicts due to funding. Selection bias may arise from excluding non-English language publications, potentially missing relevant research from other regions. Furthermore, measurement bias could be significant due to the lack of standardized testing protocols in mycelium composite characterization: different labs may use varying methods to measure seemingly identical properties. These biases should be considered when interpreting the findings, particularly the reported improvements in mechanical properties.

These limitations highlight the need for future research to establish standardized protocols for both manufacturing and testing mycelium composites, as well as the importance of comprehensive reporting

of both pre- and post-processing material properties. Future systematic reviews would benefit from a larger body of standardized research in this emerging field.

3.9. Additional considerations on material characteristics

3.9.1. Substrate particle size and morphology effects

An additional limitation of the current literature is the incomplete characterization of substrate particle size and morphology effects on heat-pressed composite performance. While particle characteristics fundamentally influence packing density, mycelial colonization patterns, and interfacial bonding, systematic investigations remain sparse, and are typically restricted to non-pressed mycelium composites, such as Houette *et al.* [34], who conducted compression and three-point bending tests comparing substrates with different particle size distributions (mean lengths of approximately 5.89 mm *versus* 2.99 mm). Their findings demonstrated a positive correlation between composite density and both Young's modulus and elastic modulus, with smaller particle sizes yielding stiffer and stronger materials. This effect can be attributed to the higher surface-area-to-volume ratio of smaller particles, which provides more sites for mycelial adhesion and creates a denser, more interconnected hyphal network. Elsacker *et al.* [8] corroborated these findings, reporting that chopped fibers with particle sizes below 5 mm significantly improved compressive mechanical properties compared to loose fibers, band concluded that mechanical performance depends more on fiber size and pre-processing than on the chemical composition of the substrate.

Beyond particle size, the morphological form of biomass fillers—specifically whether particles are predominantly fibrous, flake-like, or granular—may exert distinct effects on both pre-pressing colonization and post-pressing mechanical behavior. While no studies have systematically compared these morphological variants under heat-pressing conditions for mycelium composites, established principles from conventional composite science and wood-based panel engineering permit a preliminary conceptual analysis. The following considerations are therefore speculative and are presented as testable hypotheses to guide future experimental work.

Fibrous particles, characterized by high aspect ratios, are analogous to short-fiber reinforcements in conventional composite engineering. During heat pressing, elongated fibers could plausibly reorient perpendicular to the pressing direction, creating a layered, quasi-laminar architecture that would be expected to enhance in-plane tensile and flexural properties through fiber bridging and pull-out mechanisms. Fibrous morphologies would also offer extended surface pathways for hyphal growth along the fiber axis, potentially yielding more continuous mycelium-fiber interfaces. However, the high aspect ratio of such particles may also impede efficient packing, creating residual voids that resist closure even under elevated pressures and reduce the effectiveness of densification. Notably, the exceptional flexural strength (37.64 MPa) achieved by Song *et al.* [18] with Salix psammophila—a substrate with naturally elongated, fibrous characteristics—may in part reflect favorable stress-transfer geometry, though this hypothesis remains untested against controlled morphological comparisons.

Flake-like particles, by contrast, present large planar surface areas that could theoretically maximize contact zones for lignin-mediated bonding during thermal processing. When lignin softens above its glass transition temperature, the broad, flat surfaces of flake-like particles would be expected to facilitate the formation of extensive adhesive films, potentially yielding stronger interfacial bonds

per particle. Under compressive loading during pressing, flake-like particles would tend to align into stacked, parallel arrangements, producing a dense, layered microstructure. However, by analogy with oriented strand board (OSB) and similar engineered wood products, this preferential orientation may introduce anisotropy and create weak interlaminar planes susceptible to delamination under shear or through-thickness loading.

Granular particles, with low aspect ratios approaching unity, would be expected to offer the most efficient packing geometry, enabling high initial bulk densities prior to pressing. This close-packing behavior could favor uniform densification and may reduce the pressure thresholds required to eliminate porosity. However, the relatively low surface-area-to-volume ratio of granular particles would limit the available interface for both mycelial adhesion during growth and lignin wetting during pressing, potentially resulting in weaker particle-matrix bonding. Stress transfer in granular-filled composites would rely more heavily on the continuous matrix phase (lignin and mycelium) rather than on particle-level reinforcement, which may limit ultimate strength despite high density.

These morphological considerations would also interact with mycelial colonization dynamics in ways that remain to be characterized. Fibrous substrates may channel hyphal growth along the fiber axis, producing anisotropic networks, while granular substrates could promote more isotropic, three-dimensional hyphal architectures. Colonization rates may also differ: the interstitial spaces between loosely packed fibers would provide oxygen access and physical volume for hyphal extension, whereas tightly packed granular substrates may restrict gas exchange and limit colonization depth. Sun *et al.* [38] noted that substrate particle arrangement (loose *versus* ordered) influences the resulting pore structure and mechanical behavior, but did not extend this analysis to morphological variants under heat-pressing conditions.

Given the fundamental role of particle morphology in governing both biological colonization and thermomechanical densification, and the absence of direct experimental evidence in the mycelium composite literature, this represents a significant knowledge gap. Future research should employ controlled morphological comparisons—using the same substrate species processed into fibrous, flake-like, and granular forms—under identical pressing conditions to test the hypotheses outlined above and isolate morphological effects from compositional variables.

3.9.2. Functional properties and environmental durability

While this review focuses on mechanical properties as the primary barrier to structural andsemi-structural applications, the practical deployment of heat-pressed mycelium composites depends equally on a suite of functional properties that govern applicability and service life in construction, packaging, and engineering contexts. Although these properties are not the central focus of this review, the available literature permits a preliminary assessment of the current state of knowledge and identification of critical research gaps.

Water Resistance: Moisture sensitivity presents a particular challenge for heat-pressed mycelium composites. As discussed in Section 3.5.4, Chen *et al.* [6] demonstrated that heat pressing at 160 ℃ denatures surface hydrophobin proteins—the natural water-repellent compounds on fungal cell surfaces—reducing water contact angles from > 130 °(highly hydrophobic) to < 30 °(hydrophilic). This finding suggests a fundamental trade-off: while heat pressing dramatically improves mechanical performance, it simultaneously compromises the material's intrinsic water resistance. Shakir *et al.* [30] reported water absorption of 112.1% and thickness swelling of 126.33% for heat-pressed Rhizopus

oligosporus composites, underscoring the severity of this limitation. By contrast, Song *et al.* [18] achieved more moderate water absorption (48.1%) and thickness swelling (23.01%) through optimized semi-wet processing, suggesting that careful parameter selection may partially mitigate moisture susceptibility. For commercial applications, secondary treatments, protective coatings, or sandwich constructions with moisture-resistant veneers would likely be required. Given the importance of dimensional stability for building panels and furniture applications, future studies should systematically report water absorption and thickness swelling alongside mechanical properties using ASTM D1037 protocols to enable cross-study comparison.

Fire Resistance: The fire performance of unpressed mycelium composites has been characterized favorably in several studies, though heat-pressed variants remain largely uninvestigated. Jones *et al.* [44] demonstrated that mycelium-biomass composites exhibit significantly lower heat release rates, carbon dioxide release, and smoke production compared to synthetic polymers such as extruded polystyrene. The chitin component of fungal cell walls possesses inherent thermal stability and flame-retardant properties, forming a protective char layer when exposed to fire or radiant heat that slows heat transfer and prevents combustible volatiles from reaching the combustion zone. Chulikavit *et al.* [45] further showed that alkaline deacetylation of mycelium enhances its high-temperature thermal stability and char formation, suggesting pathways for engineering improved fire resistance. However, no studies have systematically evaluated fire performance specifically for heat-pressed materials, where the reduced porosity and altered microstructure may affect combustion behavior differently than in foam-like composites. On one hand, the elimination of open-cell porosity through densification could reduce oxygen permeability and slow flame propagation; on the other hand, the higher density concentrates combustible mass per unit volume. Given that many target applications (building panels, insulation, furniture) require compliance with fire safety standards such as EN 13501 or ASTM E84, characterizing the fire behavior of heat-pressed mycelium composites represents a critical research gap that must be addressed before commercial deployment.

Antibacterial Properties and Biological Durability: Resistance to microbial degradation and potential antibacterial functionality remain almost entirely unexplored in heat-pressed mycelium materials. While the heat-pressing process deactivates the fungal organism, preventing further growth, it does not necessarily confer resistance to degradation by environmental bacteria or other fungi. Ding *et al.* [43] recently demonstrated that chitin-containing spent mushroom substrates can impart antimicrobial properties to mycelium biocomposites, reducing colonization by certain bacterial species, but this work focused on specific formulations rather than generalizable principles. For applications in construction or packaging where biological durability is essential, these properties require systematic investigation. Commercial deployment would likely necessitate either inherent resistance mechanisms or protective surface treatments to ensure adequate service life.

However, it is worth noting that biodegradability represents one of the principal value propositions of mycelium-based materials within a circular economy framework. There exists an inherent tension between engineering resistance to biological degradation and preserving the material's capacity for natural decomposition at end-of-life. Rather than pursuing treatments that compromise biodegradability to extend service life indefinitely, it may be more strategically appropriate to match mycelium composites to applications where moderate service lifespans are acceptable—such as furniture, interior paneling, temporary installations, and packaging—rather than adapting them to permanent structural

applications where multi-decade durability is essential. This approach would align material characteristics with application requirements while preserving the environmental benefits that distinguish mycelium composites from conventional alternatives.

Collectively, these functional properties—water resistance, fire performance, and biological durability—interact with the mechanical properties discussed throughout this review to define the overall fitness of heat-pressed mycelium composites for specific end-use applications. While mechanical performance must reach threshold levels before other properties become practically relevant, a holistic material qualification framework that integrates mechanical, hygrothermal, fire, and biological performance metrics is essential for translating laboratory results into commercial products. The development of such integrated testing protocols, potentially modeled on the multi-property evaluation standards used for conventional wood-based panels (e.g., EN 312 for particleboard), represents a productive direction for future research.

4. Conclusion

This integrative review of heat pressing effects on mycelium composites reveals both promising opportunities and critical challenges in the field. While this research suggests substantial potential improvements in mechanical properties through heat pressing, the diversity in methodological approaches provides valuable insights beyond simple performance metrics.

The thermochemical analysis presented herein demonstrates that heat pressing is not merely a compaction technique but a complex thermochemical activation process that leverages the glass transition of lignin ($T_g \approx 130–150$ ℃ at 30% moisture), the thermal stability of chitin, and the cross-linking power of the Maillard reaction. Understanding these mechanisms provides a theoretical foundation for process optimization. Correlation analysis across studies revealed that pressure exerted a stronger influence on mechanical properties than temperature, suggesting that densification effects may be as important as thermal crosslinking in determining final material performance.

The most promising finding for industrial applications is that significant material improvements can be achieved with relatively short processing times—as brief as 3 minutes in some studies [36]. This suggests heat pressing could be efficiently integrated into existing manufacturing processes without causing major production bottlenecks. However, the relationship between processing time and material properties remains complex, with contradictory findings between studies.

Recent advances have demonstrated multiple pathways to high-performance mycelium composites. Ultra-high-pressure processing [6] achieved tensile strengths exceeding 12 MPa—comparable to engineering plastics—though such approaches require specialized equipment and may sacrifice the hydrophobicity of the material. Semi-wet hot-pressing with controlled moisture content [18] achieved flexural strengths of 37.64 MPa through systematic parameter optimization, validating the critical role of lignin plasticization in achieving superior mechanical performance.

The significant improvements in mechanical properties achieved through heat pressing suggest this technique could be key to expanding mycelium composites into more demanding applications. However, this review also highlights critical gaps in the literature: only two studies provided paired unpressed/pressed comparisons, and co-reporting of different mechanical properties was inconsistent across studies. While further research is needed to fully understand and optimize the process, the existing

evidence indicates heat pressing could help bridge the gap between current mycelium composite capabilities and the requirements of conventional building materials.

Key challenges remain, including the energy-sustainability paradox (heating multi-ton hydraulic presses requires significant energy that may negate the carbon benefits of biological growth), the loss of hydrophobicity upon high-temperature processing, and industrial scalability. Lifecycle assessments comparing the energy cost of pressing mycelium *versus* producing synthetic resins for MDF are urgently needed.

The challenge now lies not just in improving performance, but in developing practical, scalable processes that can deliver consistent results in industrial settings. The adoption of standardized testing protocols (ASTM D1037, ISO 16929) and the development of genetic engineering approaches to optimize fungal chitin content represent promising future directions. The transition from "growing shapes" to "engineering matter" is underway; the next phase is industrial scaling.

Data availability statement

The data supporting the findings of this study are available within the article and its references. All analyzed data from published studies are cited appropriately in the bibliography.

Declaration of generative AI and AI-assisted technologies

During the preparation of this manuscript, the authors used generative AI tools only to improve language and readability. The authors take full responsibility for the content of the manuscript. Conceptual illustrations (Figures 1 and 2) incorporate elements generated using OpenAI's image generation tools (DALL-E), which were subsequently manually composed and edited in image editing software to ensure accuracy of scientific content and text correctness. These AI-generated elements serve solely as conceptual illustrations to aid reader comprehension of composite processing concepts; they do not depict actual experimental data, micrographs, or quantitative results. The authors confirm that the use of AI-generated imagery in this manuscript complies with the journal's editorial policy on the use of artificial intelligence in academic publications. All quantitative figures, including the PRISMA diagram (Figure 5), correlation matrix (Figure 6), co-reporting matrix (Figure 7), parameter space coverage (Figure 8), and property heatmaps (Figure 9), were generated programmatically using Python's Matplotlib and Seaborn visualization libraries based on the extracted dataset.

Authors' contribution

Conceptualization, L.E.P.A. and L.A.C.; methodology, L.E.P.A. and L.A.C.; software, L.E.P.A.; validation, L.E.P.A., L.A.C. and L.H.; formal analysis, L.E.P.A.; investigation, L.E.P.A. and L.A.C.; data curation, L.A.C. and L.H.; writing—original draft preparation, L.E.P.A.; writing—review and editing, L.A.C.; supervision, L.H.; project administration, L.A.C. All authors have read and agreed to the published version of the manuscript.

Conflicts of interest

Luiz Eduardo Piá de Andrade founded a company that developed mycelium materials in 2022. It has since been closed. The authors declare no conflict of interest.

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