FUNGAL PARAMETRICS

DESIGNING A LIVING MATERIAL THROUGH BIO-DIGITAL FABRICATION

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Creative Practice PhD in Architecture

FUNGAL PARAMETRICS: DESIGNING A LIVING MATERIAL THROUGH BIO-DIGITAL FABRICATION

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ABSTRACT

Designing with living materials, as a burgeoning approach in the field of architecture, requires the development of novel design strategies and fabrication methods. Living cells have the ability to perceive and respond to environmental stimuli and this is an indicator of their intelligence, along with the capacity to form themselves. This thesis raises the question of how designers can guide the growth of a living material that has such tendencies, allowing its inherent capacities to inform the design process. The objective is to develop a framework for fabricating living materials using digital tools, with an attempt to generate new processes and practices around growing organisms. To achieve this, this thesis positions the work within the broader context of New Materialism, helping the emergence of new ideas, while adopting a parametric design approach in the context of biodigital fabrication. Furthrmore, by employing the “Research through Design” methodology, this thesis conducts conceptual experiments using fungi as a biomaterial probe to explore concepts, and to generate and assess new design strategies.

The design experiments illustrate the correlation between environmental growth parameters and morphological changes in a living material (specifically fungi in both mycelium and mushroom form). Developing input-output relationships helps in exploring philosophies and concepts, such as plasticity, linearity, and predictability. The experiments raise questions about the extent to which biological systems and their corresponding input-output relations can be considered parametric. If, indeed, they exhibit parametric characteristics, the subsequent question involves understanding the nature of these parametric systems and evaluating whether designers can employ a parametric design method when working with living materials exhibiting nonlinear behaviour.

Answering these questions leads to fabricating living materials by harnessing their developmental plasticity to give form by adjusting the environmental parameters that influence biological growth. Understanding how designers can impact organism growth without direct physical intervention leads to the development of a probability space method as a predictive tool. This method informs designers about how to manipulate input variables to achieve the desired morphological outcome. The method that has evolved through this research, based on predictions of input-output relations, may also apply to other types of biological systems, allowing meaningful interactions between the designer, organism, tools, and form. This interaction suggests that when designing collaboratively with living systems, the architect’s role transforms, requiring the relinquishment of control over the material. This, in turn, requires a new set of design processes to emerge.

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Bu tezi, egitim hayatimin son diplomasini, canim anneanneme armagan ediyorum.

Bak dedigin gibi dayandim ve 5 yilin sonunda bitti. Keske sen de gorebilseydin…

Anne ve baba sizin gibi ebeveynlere sahip oldugum icin kendimi ne kadar sansli hissettigimi bilemezsiniz. Hakkinizi ne yapsam odeyemem, sizin icin ne kadar guzel soz soylesem az kalir. Canim kardesim Zeloscugum, bu hic bitmeyecekmis gibi gelen zorlu sureci sen olmadan tamamlayamazdim. Ozkan'lar bu diplomayi hakediyor. Butun ailem, Sukran teyzem, tum destekleriniz ve benim guzel ailem oldugunuz icin cok tesekkur ederim.

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PROLOGUE

The field of synthetic biology and genetic engineering has garnered significant attention and interest in recent years. The ability to design and engineer organisms is akin to being a designer of living systems. This ability to grow organisms with specific properties and functions is not unlike the selective breeding of plants and animals, where traits are selected and cultivated over generations to produce a desired outcome. The motivation for this thesis stems from a strong personal interest and fascination with this exciting area of research. I have been greatly influenced by the radical new works emerging in art and design and this curiosity drives me to explore how architecture intersects with the exciting potential of synthetic biology and genetic engineering.

The ground-breaking works of Eduardo Kac with Alba, the genetically modified fluorescent rabbit (Eduardo 2020), and Stelarc with the third ear implant (Stelarc n.d.), have challenged traditional notions of art, and raised ethical and philosophical questions about the manipulation of living matter. These living art pieces offer a unique aesthetic experience that is both familiar and foreign, evoking a sense of fascination and unease. Not only does the artwork use living organisms, but various works that draw inspiration from biological forms and processes to create sculptures and installations (such as Patricia Piccinini, Lorenzo Oggiano, and David Altmejd, as seen in Figure 1, 2 and 3), also challenged my perception of the natural world. Through their art, these individuals explore themes such as evolution, genetic engineering, cultivation, and the relationship between humans and the environment. Their works often blur the boundaries between the living and non-living matter, offering a unique perspective on the intersections between art, science, and technology.

Figure 1

"Graham" by Patricia Piccinini depicts a surreal humanoid sculpture envisioned as the ultimate survivor on our roads.

As someone with a background in architecture, such work, with its strange, speculative, and hybrid aesthetics, has sparked a curiosity in me concerning the potential of living systems in the field of architecture. The idea of designing living systems or designing with them raised these questions in me:

- Can living organisms be programmed to grow buildings in desired forms? Is it possible to program organisms through synthetic biology in a manner similar to designing digital objects?
- What impact does using living matter have on the design of architectural structures? How will living materials behave over time?
- What would the aesthetic of architecture created using living organisms be?
- How can living organisms be utilised to create new forms of design and architecture?
- What becomes of the new roles of biology and technology in shaping the future of architecture?

Biology has already been on the radar of architects for many decades, with biological systems being incorporated into the form-finding process as a morphogenetic approach in architecture (Mertins 2007). According to architectural theorist Mertins, the morphogenetic approach in architecture, as a design method, is influenced by growth processes and developmental biology. It aims to create designs that are self-organising and adaptive, allowing for the form to emerge through interactions and processes rather than being prescribed by a predetermined plan. In this approach, design is seen as a dynamic, evolutionary process, rather than a static, preconceived form. The design process is guided by parameters and

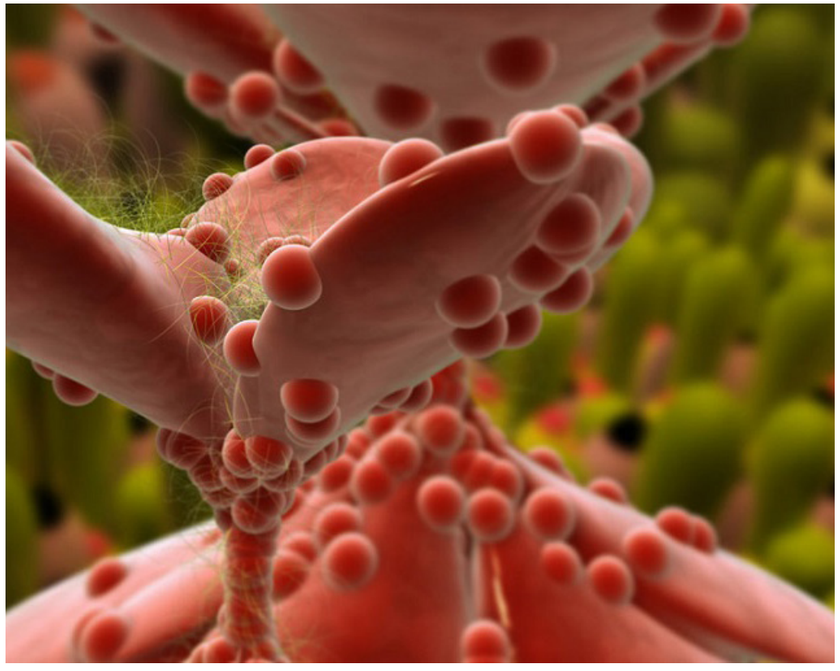

Figure 2

Quasi-Objects by Lorenzo Oggiano explores synthetic evolution, stimulating thought on techno-biological evolution.

Figure 3

The flux and the puddle by David Altmejd challenges perception through an immersive fusion of science, curiosity, and surrealism.

constraints, but the final form of the structure emerges through a series of iterations and interactions.

One other key feature of this morphogenetic approach is its use of computational tools and algorithms to model and simulate the design process. These tools allow architects and designers to (i) explore a vast number of potential design solutions; (ii) test and refine their designs; and (iii) ultimately arrive at an optimised final form. Lars Spuybroek (Spuybroek 2004), Greg Lynn(Lynn 1999), and Karl Chu (Chu 2006) can be seen as pioneers of this approach.

My motivation for this PhD was not to functionalise a biomaterial. It was more of an exploration of how we, as designers, can work with living materials and how working with a biological system could contribute to architectural design. In addition to this, my intention was to apply an existing digital design and fabrication logic, as a method, for working with living systems, leveraging my familiarity with digital methods.

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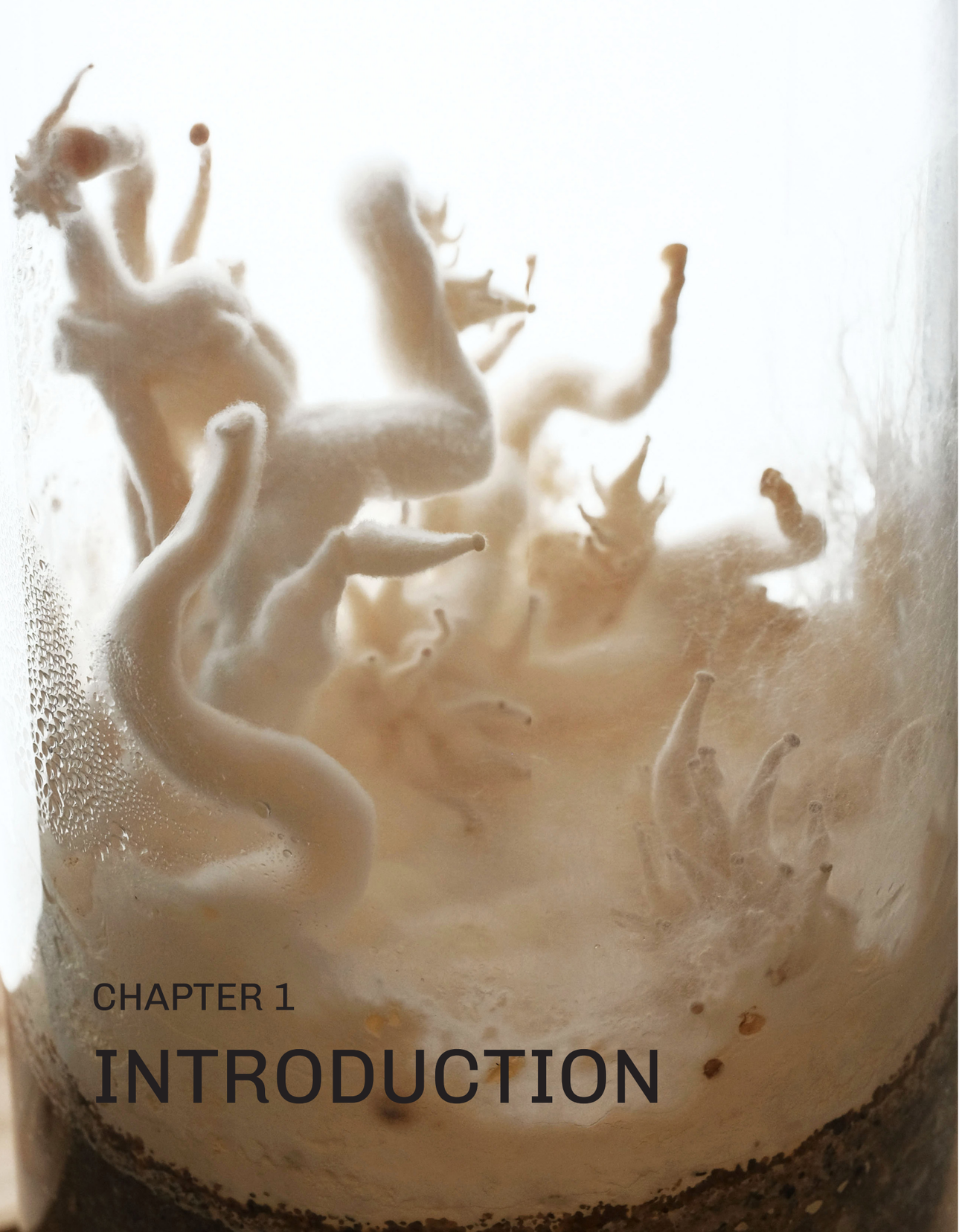


CHAPTER 1

INTRODUCTION

1. Introduction

The aim of this thesis is to examine a novel approach to the design and fabrication of a living material, specifically fungi. Fungal materials, in mycelium form, which are produced by growing the complex network of fungal hyphae (Camere and Karana 2018), have captured the interest of an increasing number of designers and architects, leading to the development of new design strategies and methods. Although this research is not intended to produce a finished product or a building material using fungi, it seeks to understand how we, as designers, can guide the growth of a living material that has certain tendencies, and allow its inherent capacities and characteristics to inform the design process. This research aims to establish a framework for design processes and fabrication methods that are centred around growing organisms using parametrically controlled digital tools.

In order to achieve this, the thesis will (i) place the work into the broader theoretical context of New Materialism which helps new ideas to emerge; (ii) look at the concepts of biodesign, biomaterials and living materials, particularly to explore different design and fabrication strategies; (iii) understand the difference between the generation of digital forms in architectural form-finding processes and the generation of biological forms in biodesign processes, as a source of inspiration and guidance; and (iv) adopt a parametric design approach, which is an established architectural design methodology, in the context of biodigital fabrication, and apply it to guide the growth of living materials. In order to do that, this thesis predominantly employs uses mixed methods methodology to generate and test new design strategies using a living material. It conducts experiments, as a proof of concept, with fungi in the mycelium and fruiting body forms, demonstrating relationships between physical and chemical growth parameters and morphological outputs. The design exploration draws upon the existing tools and philosophies within the field of digital architecture. These conceptual design experiments that focus on fungi as a non-linear material with limited developmental plasticity, consider it a challenging yet promising source for the architectural form-making process. In conclusion, this research proposes a method to inform designers about how to manipulate the input variables to achieve desired material outcomes based on predictions.

Through the translation of thought experiments into physical manifestations, this research eventually offers a biodigital fabrication and design method for living materials. The introduced method, based on input-output relations in a predictive way, may also be valid for other sorts of biological systems, as well as allowing interactions among the designer, organism, tools, and form. This interaction then indicates that when collaboratively designing with living systems, the architect's role undergoes a shift, with them granting more authority to the material. This shift then requires a new set of design processes to emerge.

2. Terms and Definitions

The incorporation of biology into architecture and design has evolved beyond just serving as inspiration for forms and mechanisms. **Biomaterials** produced by cultured biological organisms, such as plants, bacteria, fungi, and various other organisms, are used as an alternative to manufactured materials in the field of biodesign (Gazit 2016). **Biodesign** is the process of using **biological systems**, which are both single-celled and multicellular organisms, as the components of the design of products, systems, or technologies (Myers and Antonelli 2012, Dade-Robertson 2021). Biodesign integrates biological systems into designed solutions and does not just emulate nature as biomimicry does (Myers and Antonelli 2012). Instead, it integrates design with natural systems (Atzmon 2023) and uses living matter (or biomaterials) to design with (Goidea, Floudas, and Andréen 2022).

The terms “living material”, “biomaterial” and “bio-based materials” are sometimes used interchangeably, but they have slightly different meanings. Although biomaterials are cultivated from living organisms (Morrow, Bridgens, and Mackenzie 2023), they are not necessarily alive. As a comprehensive term, they refer to materials with metabolic activity, whether alive, dormant or dead. **Bio-based materials**, also known as **biomaterials**, are inherently transformative, evolving through time and across processes, in contrast to manufactured materials, which are supposed to be homogeneous, static, and stable, permitting certification and ensuring longevity (Ramsgaard Thomsen and Tamke 2022). Bio-based materials have complex heterogeneity, and unpredictable behaviours (known as nonlinearity[1]) (Ramsgaard Thomsen and Tamke 2022). They are shaped by growth cycles which then can be harvested (like timber), designed [as biopolymers or regenerated biomaterials, such as chitosan, cellulose, or sodium alginate (Mogas-Soldevila and Oxman 2015)] or still living (Ramsgaard Thomsen and Tamke 2022). A **living material** (the focus of this research) is alive and active in metabolism. Despite their tendency to exhibit non-linear behaviours, which make them difficult to work with, living materials have unique qualities that make them valuable for use. They can grow, reproduce, die, and contribute distinctive properties, such as self-assembling, self-healing, self-sustainability, and adaptability.

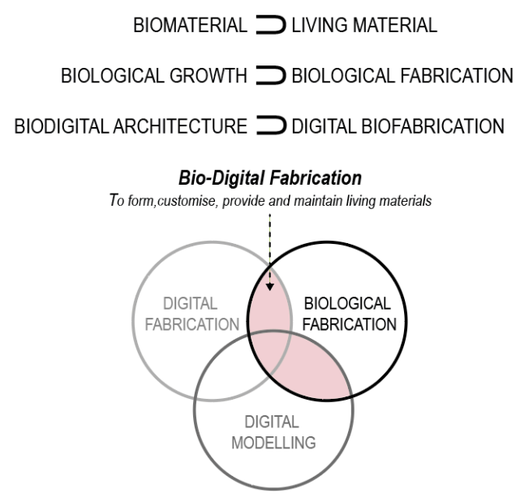
While all living materials may be referred to as biomaterials, not all biomaterials are living materials. Due to the dynamic nature of organisms, the state of living or non-living can change based on various factors. For example, a cut stem from a plant may still be considered a biomaterial, but not a living material. However, if it can regenerate and grow roots in the soil, it can become a living material again. This highlights the importance of continuously reassessing the state of materials in biodesign and considering their potential for growth and regeneration. Holding the dual state of being a living plant and non-living material blurs the line between living and death for some organisms.

To integrate living materials into design, we need to understand how designers may influence the growth and lifespan of the organism through time (Ramsgaard Thomsen and Tamke 2022). The process of designing with living matter involves a complex interplay between the designer’s intentions, the constraints and affordances of the living materials, and the systems being used. The resulting end product becomes a manifestation of this entanglement, with both human and non-human agencies playing a role in shaping the outcome. Human designers play a crucial role in determining the form and properties into which living organisms will grow. However, it is important to note that our understanding of the potential forms and properties of living organisms is still in its early stages. In the design field, the process of growing biomaterials or living materials is referred to as **biological fabrication** (or **biofabrication**), which combines the principles of design and engineering with the unique properties and behaviours of living systems (Zolotovsky 2017). It is the process of producing complex materials and artefacts through the growth of living organisms and cells, molecules, extracellular matrices, and biomaterials (Camere and Karana 2018, Mironov et al. 2009). Biological fabrication is more specific than biological growth, as it involves the deliberate growth of an organism increasing in size, mass, or complexity using engineering and manufacturing techniques for predefined goals. Biofabrication processes are coordinated with the natural processes of growth to create complex structures that have the intelligence, specificity, and active qualities of “naturally developed” entities, rather than simply manufacturing with biological or living building blocks (Rotondi 2023). For example, mycelium composites, created by growing the intricate network of fungal hyphae over a substrate, are biologically fabricated materials (Camere and Karana 2018), whereas wood is not viewed as a product of biofabrication. Although wood is a biological material, it grows naturally without human intervention, combining design and engineering principles.

Biofabrication processes employ various strategies and techniques to produce materials with desired properties and functionalities, using living cells. The integration of novel fabrication technologies plays a crucial role in advancing biofabrication methods, as the organism has needs and requirements during the fabrication process while growing and making the final object (Andréen and Goidea 2022). With a form-based approach, computational

[1] *A detailed explanation will be provided in Chapter 6.*

Figure 1
Diagram presenting the relationship diagram between sub-cluster and top-cluster. “⊃“ indicates superset meaning one set contains all the elements of another set.



principles, digital modelling, fabrication tools, and methods originally used in architecture have also been adapted as tools to fabricate living systems in the field of biodesign. This approach, known as “digital biofabrication” and “biodigital architecture or practices” combines biological principles with digital design and fabrication methodologies. **Digital fabrication** of living materials using methods from architectural manufacturing is referred to as a digital **biofabrication** approach (Camere and Karana 2018). Digital fabrication technologies utilise manufacturing equipment that is directly driven by digital data to create diverse part geometries (Cutieru 2020). The design information originates in digital form, is transferred to computer-aided manufacturing software, and is then directed to a specific machine, such as a 3D printer or CNC milling machine (Formlabs, n.d.). While these technologies are continuously evolving, they use one or more of the following manufacturing techniques: additive, subtractive, and formative manufacturing or robotic manipulation in any form (Keating and Oxman 2013). The digital biofabrication approach also integrates biological tools with advanced computer technologies, and leverages digital manufacturing tools to enable precise control over the cell (as living material) distribution (Jauk et al. 2021). This then leads to scalability, reproducibility, and automation of biological processes (Bitting et al. 2022). Examples of digital biofabrication include 3D-printing cells and using algorithms to guide cell distribution, as well as cultivating cells in digitally controlled growth chambers or bioreactors.

Biodigital architecture is a broader, more conceptual term, used to encompass Artificial Intelligence, Algorithmic and Parametric Approaches, Machine Learning, and Computation (Estévez and Navarro 2017). It refers to the combination of biological and digital architectural processes, combining the intelligence derived from nature with computational-based technology (Estévez and Navarro 2017). While there is an overlap between the terms “digital biofabrication” and “biodigital architecture”, digital biofabrication focuses more on the technical processes and tools used to manipulate biological materials, while biodigital architecture takes a more holistic and conceptual approach, considering how these techniques can be applied in the context of architectural design (Figure 1).

This thesis uses the term **biodigital fabrication**, which not only refers to the use of digital fabrication systems, but also encompasses computational thinking to guide the growth of living materials for design purposes. Biodigital fabrication includes a range of techniques, tools, and purposes that can be employed in diverse ways to fabricate living systems. It can be categorised into three different approaches:

I. Use of digital tools to generate forms through moulds, scaffolds or nutrients that living materials follow/grow on/in

The form and structure of living materials can be designed using digital modelling and fabrication tools, for the creation of scaffolds or templates on which the materials grow. Predefined digital data plays a key role in informing the precise and customizable forms of living materials (Abdallah and Estévez 2021). Examples of this approach include the “Mycelium Chair” by Eric Klarenbeek (Fairs 2013), or Natalie Alima’s “Bio Scaffolds” (Alima, Snooks, and McCormack 2021), where a 3D-printed, degradable scaffold is utilised to hold the organism and

Figure 2
Photographs of 3D-printed scaffold by Natalie Alima (left), substrates by Fraunhofer (centre) and pre-inoculated substrates by Ana Goidea et al. (right) (Bitting et al. 2022).

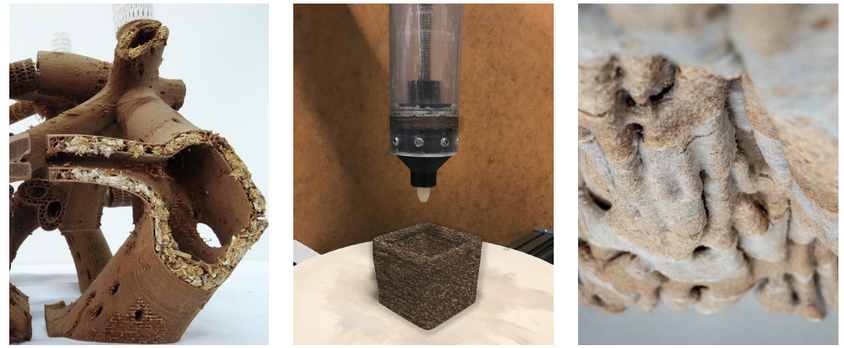


Figure 3
Diagrams of Hybrid Living Materials (left) (Smith et al. 2020), Living Manufacture (right) (Arnardottir et al. 2022) projects.

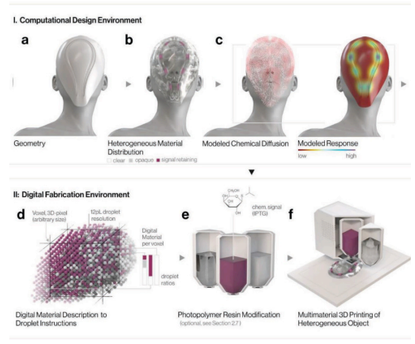


Figure 4
Paludarium Osamu (left) (Makoto 2017), Elowan (centre) (Sareen and Maes 2020), Flora Robotica (right) (Hamann et al. 2017).

its substrates. Similarly, 3D-printed substrates can be inoculated with the organism, such as fungus or bacterial cellulose, after the printing process is completed as in “FungiFracturing” by Fraunhofer (Krayer M.A., n.d.). Another example is the “Protomycokion Column” by Ana Goidea et al., where 3D-printed scaffolds consist of pre-inoculated living mycelium mixed with substrates (Andréen and Goidea 2022), also known as bio-inks (Bitting et al. 2022) and bio-printing. These examples demonstrate the application of additive manufacturing methods in creating scaffolds or surfaces on which living materials can grow (Figure 2).

II. Use of digital tools to customise the growth and behaviour of living materials

This approach involves the use of digital systems to influence and modulate the behaviour of a living material during its fabrication/growth, using bespoke computational algorithms (Camere and Karana 2017). It merges biological and digital computation so that living materials can be fabricated for specialised functions (Rotondi 2023). For example, the “Hybrid Living Materials” project by the Mediated Matter Group demonstrated a method of integrating 3D digital design and printing with engineered bacteria, which then allowed them to achieve programmable, replicable control of gene expression across the surface of 3D-printed objects (Smith et al. 2020). Similarly, the Living Manufacture project aims to develop an Engineered Living Material[2] (EML) based system to produce Bacterial Cellulose (BC) pellicles using fermentation vessels and digital fabrication methods. These BC pellicles can respond to different stimuli and be patterned in three dimensions (Arnardottir et al. 2022). This approach highlights how the different sets of properties of a living material can be precisely and intentionally controlled digitally, leading to new material properties (Figure 3).

III. Use of digital tools to provide certain growth conditions for living materials

Digital fabrication technologies can be utilised to programme living material’s behaviour by calibrating the nature of the physical and chemical environment. They help to guide the growth of an organism by automating, regulating, and facilitating habitats (Karana, Barati, and Giaccardi 2020). The use of robot arms, drones, monitoring systems, growth chambers, incubators, and bioreactors provides environmental regulation and maintenance of resources, including water, light, or nutrients. This leads to minimal direct physical contact between human and non-human and optimises the growth of organisms from single cells through to harvest. In a sense, the digital tools function, and are programmed, as gardeners. For example, Azuma Makoto’s “Paludarium Osamu” incubator is an automated, encapsulated, environmental ecosystem that grows a bonsai tree by digitally controlling and regulating water, soil, air, and light quality (Makoto 2017). The incubator works like an “automated home” in which the organism can complete its growth stages and lifecycle in a succinct and condensed way. The Elowan robot, developed by MIT Media Lab, has similar goals to the Paludarium Osamu, which is to satisfy the needs of their plants (Sareen and Maes 2020). Elowan, as a mobile platform carrying a plant, rolls itself towards a nearby light source if needed, responding to the stimulus gathered through the electrodes attached to the stems and ground of its plant (Sareen and Maes 2020). Another organism-robot interaction[3] is the

[2] ELM is made of live cells and biomolecules like proteins or DNA that either construct or assemble the material itself or modify its functional performance in some ways (Nguyen et al. 2018).

[3] This interaction can also be called as a cyborg.

‘Flora Robotica’ project, where plants and distributed robots are coupled to create architectural spaces (Hamann et al. 2017). Plants act as a scaffold to carry the robot, and robots can direct the growth of the plants by using LED lights and sensors to form pre-programmed shapes. They also measure the plants’ condition by controlling soil humidity levels and responding, for instance, by pumping water, if required (Hamann et al. 2017) (Figure 4).

Overall, biodigital fabrication refers to a range of possibilities for the creation and manipulation of living materials that can be precisely controlled and tailored to specific applications. Digital tools help to facilitate biologically active organisms, either by generating form, customising growth, or providing specific growth conditions. While the form is predefined digitally by the designer in the first category, it is generated by the living material itself with support and guidance from the designer in the second and third categories. As a result, new relationships occur between the organism, the designer, and the digital system. This interaction forms and grows organisms, converts them into a programmable material system, and functionalises them for relevant applications (Zolotovsky 2017).

In the context of this thesis, biodigital fabrication does not rely on pre-designing forms. Instead, the form emerges from the living material as a result of interactions with digital tools that consequently alter the environment, promoting the material’s growth (as in the third category outlined above). While guiding the growth of fungus as a material through environmental regulation and maintenance, it controls the input variables through a parametric design-inspired method. It attempts to tune and associate the morphological properties of fungi (as outputs) with environmental variables (as inputs). This approach seeks to apply an established digital architectural design method to living systems, distinguishing it from other examples in the third category.

3. Focus of This Research

As a rapidly developing discipline, the incorporation of living materials into the design as a biomaterial is clearly different in terms of fabricating, working with, and managing (Ramsgaard Thomsen and Tamke 2022) due to their metabolic processes. They differ from non-living materials, which are static and do not change significantly over time unless acted upon by external forces (Ramsgaard Thomsen and Tamke 2022). Manuel DeLanda presents this difference with the theory of “New Materiality” by combining the ideas of the philosopher Deleuze and the theoretical physicist Henri Poincaré. According to DeLanda, “**Obedient Materials**” simply obeys fundamental rules and derives all its abilities from the supreme laws based on Newtonian theories (DeLanda 2015). During the traditional materialisation of a design with obedient materials, the most appropriate material is typically selected, not designed, from a set of other materials (Oxman 2010). The process of material selection and design takes place in separate phases, with the chosen material becoming a building block that is later included in the design (Oxman 2010). On the other hand, the concept of “**New Materiality**” introduces the idea that matter has its inherent morphogenetic capabilities and “does not need to be commanded into generating form” (DeLanda 2015). With a New Materialist approach, the material’s abilities and capacities are integrated into the design process,

directly influencing the final product. This interpretation of materiality opens new horizons for architectural theory about how to handle living materials creatively. Creativity might entail suggesting new fabrication methods that enable new ways to direct growth, developing a better understanding of the material and its behaviour, and approaching it in a spirit of cooperation and exploration.

Adopting a New Materialist approach in the biofabrication of living materials introduces a shift towards more integrated and autonomous systems, where the material is able to grow and evolve on its own with subtle physical intervention from the designer. This shift is based on the notion that living matter has the ability to generate form on its own (Kretzer 2017). Due to their autonomy, they go beyond the role of simply shape-filling as material blocks (Oxman 2010), performing abilities such as self-assembly, self-healing, self-sustainability, and adaptability (Nguyen et al. 2018). They act as active materials with intelligence instead of obedient ones. However, it is important to note that not all currently practised digital and biological fabrication methods may allow this level of autonomy in living materials. For example, top-down fabrication strategies (stated above as the first category), involving the cultivation of cells within predefined moulds or scaffolds (Schmidt et al. 2021), can potentially constrain their growth to specific boundaries and shapes. The expression of a New Materialist approach in highly restrictive systems is limited when designers act as top-down controllers of events, rather than as co-designers of the systems (Armstrong 2016).

The restraining biofabrication methods prevent material emergence and reduce the degrees of freedom the materials have. **Material emergence** refers to the ability of a material or system to generate new qualities or behaviours that are not explicitly specified by the designer or programmer (Ratter 2012). Emergence is intrinsically tied to "**degrees of freedom**" signifying any relevant means of alterations in the system (DeLanda 2007). In methods allowing higher degrees of freedom, new relations emerge, and the format of the making tradition changes. The role of a designer, traditionally perceived as having authority over the design process and the final product, transforms when working with systems that cannot be fully controlled. This shift presents an opportunity for innovative biodesign and biofabrication processes, fundamentally altering the roles of both the designer and the material.

This thesis argues that while fabricating living materials, a designer's role should be more like that of a gardener, nurturing the organism and guiding its growth in a particular direction. It explores a design process and digital fabrication method for living materials that enables them to express their morphological tendencies and capacities for design applications. By embracing a New Materialist perspective, it tries (i) to collaborate, interact, or build a dialogue with a living material as a non-human actant during the design process; (ii) to illustrate an input-output relationship by defining the parameters that manipulate an organism's morphology; and (iii) establish a new framework for biodesign processes. This investigation applies a parametric design approach, already used in architecture, to living materials, facilitating their non-obedience, self-assembly, nonlinearity, or unpredictability. It ultimately proposes a biodigital fabrication approach demonstrated on fungi, through regulating their physical and chemical environments.

4. Research Questions, Aims and Objectives

This thesis explores a biofabrication approach that investigates the potentials and limitations of designing the growth of living materials. It sits within the context of the much wider and general question "**How can we design with the living?**" More specifically, it asks "**How can we design and fabricate a continuously metabolising living material (such as fungus), by allowing its tendencies and capacities? How can we minimise our interference in the fabrication process?**" The importance of addressing these questions lies in defining the evolving role of designers within a system where the material has agency and can influence the design outcome.

This research aims to develop a design framework and biodigital fabrication method to guide the growth of a living material (fungus) that facilitates interaction between the designer, the material, and the tools used in digital architectural practice. It attempts to enable the control of living materials in a manner that embraces a collaborative approach, rather than imposing predetermined forms or working against their natural tendencies. Providing such a tool and biodigital fabrication method also demonstrates the challenges when design interventions are applied to non-linear or non-obedient materials. To address the stated research question and the aim, it is essential to investigate and find answers to a series of objectives and related sub-research questions. The objectives of this research are:

Objective 1: To examine, understand, and redefine the relationship between a living material and design practices in the context of biodesign.

Objective 2: To develop a biofabrication tool and method that (1) is applicable to "making-through-growing[4]"; (2) allows material interaction through its developmental parameters; and (3) allows designers to work with the tendencies and capacities of living materials.

Objective 3: To develop a design concept that presents the methods for designing living materials under untested conditions.

These objectives will help to gain a deeper understanding of the unique opportunities and challenges of biodesign processes, and potentially discover new ways of approaching the design and fabrication of living systems. To accomplish these objectives, many questions need to be addressed. These questions are listed in Table 1, which illustrates the interconnectedness of the objectives, questions, and methods used to answer the questions.

The outcomes of this research will be valuable for the advancement of architectural design, biodesign, and biodigital design fields, adding existing knowledge and enhancing innovative opportunities, especially for prospective studies. Therefore, this research is expected to engage readers from architecture, design, and materials disciplines, who are interested in exploring the potential of autonomous materials.

[4] *"Making-through-growing" approach will be explained further in Chapter 3.*

Table 1

Mapping research objectives with corresponding research questions.

Objectives	Sub-Questions	Methods	Chapters
To examine, understand, and redefine the relationship between living materials and design practices in the context of biodesign.	What is the difference between making and growing in biodesign processes? What are the tendencies and capacities of living materials?	A systematic analysis of literature review through the study and analysis of different projects.	C3: New Materialism
To develop a biofabrication tool and method that: (1) is applicable to "making-through-growing", (2) allows material interaction through its developmental parameters, and (3) allows designers to work with the tendencies and capacities of living materials.	What tools and methods are applicable to making-through-growing? How can we interact with the material through its developmental parameters instead of interacting with them directly?	A systematic analysis of approaches to parametric design from literature leading to a broader philosophical understanding of design and design methods.	C4: Parametricism
	How can fungus be used as a living material to test the concept of a New Materialist biofabrication approach? What are the characteristics of fungi and what influences their growth and development? What are the constraints, if any, on the growth stages of fungi?	Early (mycelium) experiments were conducted as preliminary design experiments.	C5: Plasticity and Form Taking
	What are the most important parameters in informing fungal (fruiting body) morphology? If we develop a form of a parametric design tool for biofabrication, what does it look like and how does it work? How are the input parameters and output morphologies interrelated?	Systematic and control-based (fruiting body) experiments using a controlled environment (growth chamber).	C6: Linearity and Form Giving
To develop a design concept (based on predictions), presenting the methods for designing living materials under untested conditions.	Can we develop a design process which allows the designer to predict the form of their material for conditions that have not been tested yet?	Creation of diagrams as predictive tools, based on the insights gained from the prior chapter.	C7: The Probability Space

5. Research Methodology

To respond to the sub-questions outlined in Table 1, this thesis adopted the mixed methods methodology that combined various methodologies, methods, data sources, and research strategies (Plowright 2019). The integrated methodologies included a literature review, research through design (RtD), and both quantitative and qualitative methods. RtD entails "iteratively designing artefacts as a creative way of investigating what a potential future might be" (Zimmerman, Stolterman, and Forlizzi 2010). Designers and researchers using RtD, engage in experimentation with new materials and processes (Godin and Zahedi 2014), thereby contributing to the generation of knowledge. During this research, the following steps were taken:

I. Conducting a literature review

Literature reviews helped to understand (1) different approaches to incorporating materials in design processes; (2) the methods used for the biofabrication of living materials and how other designers have interacted with living materials; (3) the characteristics of fungi and procedures used for growing them; and (4) the conceptual architectural digital design and fabrication methods that could also apply to living systems.

II. Conducting conceptual science-led design experiments

Design experiments refer to the integration of scientific principles with design thinking to investigate and conceptualise new ideas involving fungi that helped to understand the characteristics and behaviour of living matter. This approach involved setting up experiments to gather data rigorously, analyse behaviours, and test hypotheses, to answer design-oriented questions reliably. While the integration of science and design was not new, the specific methods of crafting living materials without genetic manipulations made each experiment unique.

Fungi (both mycelium and mushrooms) were viewed as a proxy, serving as a medium to investigate the interface between biological systems and design. They offer insights into how living materials can actively participate in shaping outcomes and whether the developmental processes of living materials can be employed as a fabrication method. The mushrooms used in this design investigation symbolise adaptability, responsiveness, and the potential of living systems to inform new approaches to design and fabrication. In this context, they are not the end goal, but a means to interrogate and expand our understanding of bio-design and complex systems. They also helped to investigate the viability of implementing a parametric design approach while working with living materials.

A combination of methods was used, including the biomaterial probe method (Ramirez-Figueroa 2017) and scientific methods to gather data. These experiments also facilitated the exploration of the extent to which biological systems can be considered parametric and what kind of parametric systems they are. The iterative process of design experiments, testing, and refinement contributed to optimising the biofabrication process, ultimately leading to more successful outcomes.

III. Implementing visual documentation in design-led research

This process included the systematic capturing, measuring, and analysis of the experiment results with various bespoke methods. Since there is no standard method for measuring conceptual experiments, new methods need to be developed for this process. Tools, such as photography, microscopic imaging, and 3D scanning, played a vital role in providing a tangible record, facilitating reflection and analysis, aiding iteration and improvement, and enabling effective communication of the research process and results. After evaluating the results, a new design concept for biodigital fabrication was proposed through diagrams and other visual representation methods. This concept challenged the existing biofabrication methods by bringing an element of unpredictability and adaptability to the field of biodesign with the transformative potential of visual documentation in design-led research.

6. Chapter Summaries

The thesis comprises eight chapters. The structure follows a deliberate and incremental approach in which the outcomes of each chapter lay the groundwork for the issues to be investigated in the succeeding chapter. Each chapter progressively delves deeper into the subject matter, providing an extensive analysis of the topic and examining various facets of biodigital fabrication.

Chapter 2: Methodology

Key concepts: Mixed methods methodology, research through design, literature review, biomaterial probe method, design experiments, science-led methods, and visual representation methods.

The methodology and methods employed in this research are presented in this chapter. The chapter outlines the use of ‘mixed methods’ as the chosen methodology, integrating a literature review, RtD, and multiple quantitative and qualitative methods. RtD facilitates an iterative approach to explore and test ideas through hands-on experimentation and prototyping. The RtD approach is also well-suited for interdisciplinary research involving design and science, as it supports the integration of practical and theoretical knowledge.

The chapter also explains the selection of the organism (fungi) used to test design ideas and generate data through a science-led approach, involving growing fungi and utilising them as a material for biodigital fabrication. The methods used and developed, including material scientific methods, and visualisation methods, are described in detail to demonstrate how data was generated. After analysing the data, diagrams, and diverse representation strategies (as part of the visualisation methods) a method for designing living materials based on predictive models was suggested. Finally, the chapter summarises how these methods enabled the testing of the concept of designing with living materials and provided insights into the material’s potential for design purposes.

Chapter 3: New Materialism

Key concepts: Making and growing, components of a living system, New Materialism, form-making and form-finding, top-down and bottom-up approach, material emergence.

This thesis starts with questioning the difference between making and growing a material, as addressed in Objective 1 found in Table 1. It discusses the unique properties of biological materials which distinguish them from materials without a metabolism. Biomaterials are capable of self-assembly and developmental processes that result in a unique morphology that can be effectively utilised. By considering their tendencies and capacities, the New Materialist logic can be applied to biomaterials. This logic differs from traditional top-down form-making approaches that prioritise achieving an optimal form, without considering material emergence. Many current approaches treat biologically grown materials as non-reactive materials, despite their unique characteristics, such as in fabricating mycelium bricks. This chapter highlights the difference in the way living materials are used in design versus how they could potentially be used, given their unique properties and capacities, through several examples providing specific instances. The evidence and methodology used to demonstrate this difference originate from a systematic literature review and analysis of various projects. This chapter concludes by questioning how to work with the tendencies and capacities of biological materials with a New Materialist approach.

Chapter 4: Parametricism

Key concepts: Digital and biological form generation, biological parametricism, direct and indirect associations, design space.

The chapter questions the methods which apply to making-through-growing theoretically, as described above in Objective 2 (1). It discusses the potential of using an existing fabrication method from architecture design, allowing us to think about a designing and making process with systems that we are not fully in control of as designers. Parametric design is one such method that is highly relevant for application in biofabrication. The development of tools to comprehend and work with the tendencies and capacities of living materials is necessary, as discussed in Chapter 3. The parametric design offers a promising approach due to its systematic methodology, which involves the use of algorithms to generate designs or morphologies for digital objects. It allows interaction with the material through its developmental parameters, rather than direct contact. The chapter focuses on the application of the parametric approach, specifically to the fabrication of living materials. The methodology used involves a systematic analysis of approaches to parametric design from literature, as well as a broader philosophical understanding of design and design methods. Based on this analysis, the chapter hypothesises that a form of parametric design could be developed for biological fabrication. As a result, the next chapter sets up material experiments to test this hypothesis.

Chapter 5: Plasticity and Form Taking

Key concepts: Developmental plasticity, nature of fungus, material experiments with mycelium, form taking and form giving.

The main objective of this chapter is to explore the potential process that may be involved when using a parametric approach in biofabrication and to determine which organism would be best suited for demonstrating this system, as mentioned above in Objective 2 (2) and (3). The chapter focuses on the use of fungi, and highlights the two critical growth stages

- mycelium and fruiting bodies. Initially, the most important parameters affecting fungus morphology are defined, and a series of experiments are designed. Early design experiments using mycelium served as a method using a craft-like process of material experimentation to gather evidence.

The growth of mycelium is observed, and it is found that mycelium responds to nutrient levels and physical constraints, and it tends to take form, rather than give it. On the other hand, mushrooms exhibit a greater degree of morphological adaptability and have the ability to give form, on a macro scale. The term “developmental plasticity” is introduced to explain this difference, which refers to the relationship between phenotype and environmental factors, and suggests the level of external control for biofabrication. This chapter concludes that to evaluate the concept of directing living systems with a parametric approach based on their developmental processes, mushrooms are more suitable than mycelium. Hence, the subsequent chapter focuses on experimenting with mushrooms.

Chapter 6: Linearity and Form Giving

Key concepts: Linearity, nonlinearity, correlation, thresholds, tipping points, dominant factor, fruiting bodies, growth chamber.

This chapter explores the complex relationship between environmental parameters and the morphology of fruiting bodies, with control-based thought experiments. Studying fruiting bodies is more interesting due to the complex relationship between macro form and environmental parameters. To understand this relationship, the most influential parameters affecting fungal growth are identified, as mentioned in Objective 2 (2) and (3).

Controlled environments are then established through a growth chamber that helps to set up different conditions. The experiments provide systematic evidence informing the use of a parametric approach in biofabrication. The findings show that although the relationships between different parameters and fungal forms are nonlinear, they are correlational and predictable within upper and lower tipping points, with multiple environmental factors contributing to the same output. The chapter also acknowledges that there are variations in these relationships. However, variations are viewed as points that define parametric areas or zones. The topic of whether it is possible to create a design procedure that enables us to predict the shape of mushrooms serves as the chapter’s conclusion.

Chapter 7: Probability Space

Key concepts: Probability space method, drawings of expectations, range of tolerance.

After establishing the fundamental parameters of mushroom growth, Chapter 7 examines whether it is possible to develop a design process which allows the designer to predict the shape of a mushroom for conditions that haven’t been tested yet, as addressed in Objective 3. Due to the emergent nature of mushrooms, which involves complex processes and morphologies, there is a significant margin of error in predicting the shape of a mushroom. The chapter introduces the concept of the “probability space” to describe this uncertainty. The probability space represents the range of possible outcomes for a given set of conditions, considering variations and errors in the growth process. The chapter then discusses the use of development diagrams as a method to compare predicted outcomes with limited scientific experiments as evidence. Although the proposed initial diagrams are two-dimensional, they are utilised to speculate about the outcomes in 3D using a novel method.

Chapter 8: Conclusion

Key concepts: Limitations, potentials, future work.

The previous chapters respond to the objectives by examining and redefining the relationship between biodesign and one type of living material, developing methods for biofabrication, and proposing a predictive design concept for working with living materials. In Chapter 8, the focus is to synthesise a design concept development for a new parametric system in biodigital fabrication. The proposed system is based on the concepts of plasticity, linearity, and probability space. Parametrising living materials as a novel design concept and biofabrication method: (1) changes the role of the material, actively involving it in the design process; (2) redefines the designer’s role to design a spectrum of possible outcomes instead of predetermined forms; and (3) facilitates the use of digital tools to allow designer-material interactions and to manage the complexity inherited in living systems. Although currently limited to a subset of biology, this approach could potentially be applied to a range of different biological systems in the future. The chapter also discusses how this system could be developed for more complex biological systems and useful purposes, while also acknowledging its limitations.

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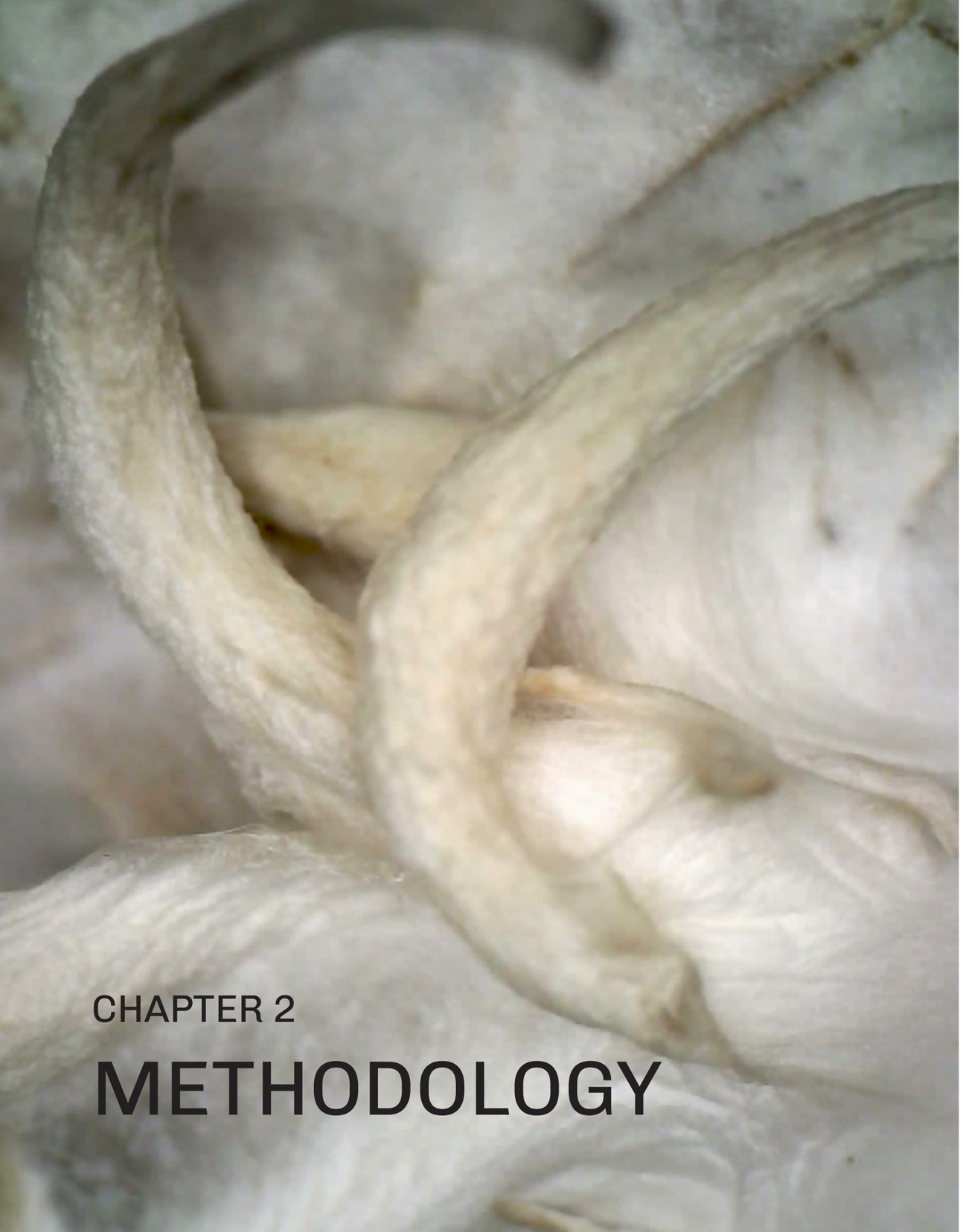

CHAPTER 2

METHODOLOGY

1. Introduction

The methodology chapter embarks on a journey to challenge the conventional approach to fabricating living materials in the context of biodesign. Rather than merely conducting a theoretical review and analysis of existing approaches, this study proposes a design approach for working with living materials that embrace their inherent growth tendencies with minimal interference. The objective is to develop knowledge in designing with living materials and to explore alternative fabrication techniques through creative practice research. Achieving these objectives is crucial because integrating living materials as non-human actants into the design process fundamentally influences the way we design. The role of the designer changes in a system where they do not have full control over the material. Instead of being a traditional maker or fabricator, the designer takes on new roles, such as a gardener, bio-crafter, guider, or system utiliser. This shift in the design process requires reconsideration of approaches and methods. Additionally, working with living systems that have flexible/changeable properties, demands a comprehensive understanding of how the material will react to varying conditions and how this response can be harnessed in the final product.

To accomplish the objectives and answer questions, as mentioned in Chapter 1, this thesis employed an **integrated methodology**, also known as **mixed methods methodology**. This methodology takes a holistic perspective on research, combining various methodologies and methods, different data, and research strategies (Plowright 2019). The integrated methodologies used in this research to investigate designing with living systems include a literature review, research through design (RtD), and both quantitative and qualitative methods (Table 1). **Literature reviews** were conducted to gather and analyse existing knowledge, and contextualise the research approach within the field. **Research through design** allowed the testing of ideas, generating data for the evaluation and creation of new arguments. The research also involved the application of multiple quantitative and qualitative methods, such as the **bio-material probe method**, and **visualisation methods** to study living materials. In this process, design experiments were used, as they helped to explore emerging design ideas (Krogh, Markussen, and Bang 2014). In those experiments, a **scientific method** was employed to analyse data and test hypotheses.

Table 1

The methodologies used in the thesis.

Objectives	Methods	Methodology
To examine, understand, and redefine the relationship between living materials and design practices in the context of biodesign. (Chapter3)	• A systematic analysis of the literature review	Mixed methods methodology
To develop a biofabrication tool and method that: (1) is applicable making-through-growing, (2) allows material interaction through its developmental parameters, and (3) allows designers to work with the tendencies and capacities of living materials. (Chapters 4, 5, and 6)	• Literature review • RtD including material experiments, biomaterial probe • Scientific methods (Systematic, control-based design experiments)	
To develop a design concept (based on predictions), presenting the methods for designing living materials under untested conditions. (Chapter 7)	• RtD including visualisation methods and diagrams	

Although the thesis might initially seem to follow a straightforward scientific methodology due to its experimental approach, it is fundamentally design research. The experiments are driven by design questions rather than scientific hypotheses. At its core, the research explores what the designer learns through the process, how the designer's interpretation of observations guides decisions, and how their reflections influence the journey. While both quantitative and qualitative observations are made during the process, the judgments and decisions are inherently subjective and personal. Decisions about the next steps are informed by quantitative measures but are ultimately shaped by personal judgment.

The primary concern of this research is not to test universal truths but to advance design practices. These characteristics qualify the process as a personal, reflective, and design-driven exploration. This aligns with a mixed-methods approach that combines Research through Design (RtD) with scientific methods, embedding knowledge in artifacts and processes rather than treating it as detached or purely analytical.

Before delving into the methods and methodology used in this research, an exploration of prior studies focused on designing with non-linear, self-assembled, and non-traditional materials was conducted. Designing within materials that provide a special challenge and over which the designer has no complete control is still an emerging field, therefore gaining insights from prior research in this domain was helpful. With this goal in mind, three PhD

Table 2

The summary of three RtD approaches.

Research	The Design	Designer's Role	Methods and Methodologies
Adam Blaney Designing Parametric Matter: Exploring adaptive material scale self-assembly through tuneable environments (Blaney 2019)	Iterative Prototyping for developing an adaptive design and fabrication system. Experiments: 1. Mineral accretion 2. 2D Generative paint 3. 3D Ink diffusion	To develop multiple prototypes which highlight how an adaptive design and fabrication system can be created when using materials that are intent and self-assembled.	RtD: Explorative and flexible design/ fabrication process enables flexibility and facilitates hands-on engagement.
Carolina Ramirez-Figueroa Biomaterial Probe: Design engagements with living systems (Ramirez-Figueroa 2017)	Evaluation and generation of design strategies: Material proxy, parametric, diagrammatic, chemical/physical state space. Experiments: 1. Synth morph 2. Bacterial crystals 3. Bacterial morphologies 4. Bacterial assemblages	To develop a strategy that enables a dialogue with biological systems, establishing its material possibilities.	The probe method: To focus on the design process rather than the design output. RtD: To show how design is transformed and redefined using living systems.
Katia Zolotovsky Guided Growth: Design and Computation of Biologically Active Materials (Zolotovsky 2017)	A design process that creates hybrid materials composed of engineered living cells and non-living scaffolds. Experiments: 1. Nanoscale: engineered cells 2. Mesoscale: keeping bacteria alive, responsive 3. Macroscale: modulate the growth environment	To demonstrate ways to program materials to sense their environment, process information, and adapt their structures, properties, and biological functions.	Mixed methods from synthetic biology, material science, architectural digital fabrication Experimental methodology: To develop new materials that embed biological responsiveness, architects need to learn to design using new mediums of materials in liquid or hydrogel phase.

research projects led by researchers with backgrounds in architecture were examined to develop an approach to the design with living materials (Table 2).

Adam Blaney (from Lancaster University) worked with ink and paint, while Carolina Ramirez (from Newcastle University) and Katia Zolotovsky (from MIT) worked with bacteria (Blaney 2019; Zolotovsky 2017; Ramirez-Figueroa 2017). By exploring these unique materials, the researchers are pioneering new ways of working with their fabrication techniques and pushing the boundaries of what is possible. Their work is important, as it has the potential to lead to innovations and solutions in the field of design and architecture. As their research makes up a range of methods, it involves a certain degree of trial and error. They explore new methods that often require experimentation and iteration to refine and improve their approach. By moving forward through trial and error, the researchers are able to identify what works and what doesn't, allowing them to make informed decisions and progress towards their goals. In this way, these attempts are an important part of the research process, as they allow for exploration, learning, and growth, and ultimately lead to new insights and discoveries.
Blaney, Ramirez, and Zolotovsky ran a series of consecutive design experiments to explore and refine their fabrication techniques. During this process, they likely developed their unique methods for setting up and carrying out the experiments, and developing methods for measuring, recording, and analysing the results. Their methodology embraced mixed methods, necessitating the integration of scientific observations and measurement methods with design studies. As a result, they contributed to the literature by building a comprehensive understanding of their materials and developing new, innovative approaches to working with them.

This thesis used an integrated methodology through the combination of RtD, literature review and other methods (Table 3). It started by examining the distinctions between making

Table 3

The summary of the RtD approach used in this thesis.

Research	The Design	Designer's Role	Methodologies
Dilan Ozkan Fungal Parametrics: Designing a Living Material through Biodigital Fabrication	Developing a biodigital fabrication method to guide the morphology of living materials with minimal physical intervention. Experiments: 1. Guiding mycelium 2. Guiding mushrooms 3. Designing mushrooms by using prediction methods	To explore a biodesign and fabrication approach for living materials which exhibit nonlinear behaviour.	Mixed method methodology: a combination of literature review, RtD, and hybrid methods including biomaterial probe method, scientific methods, and visualisation methods.

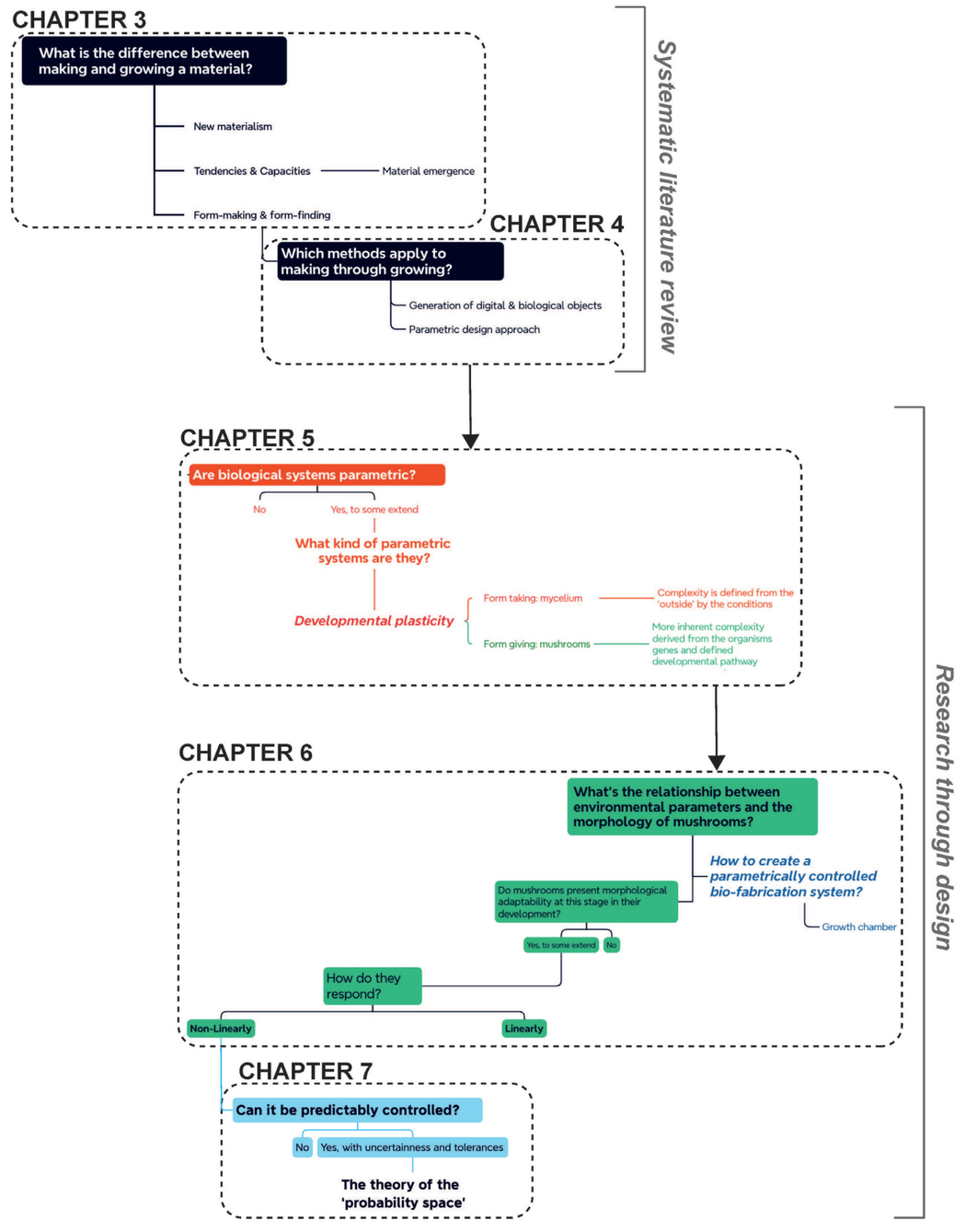

Figure 1

Diagram illustrating the thesis structure.

and growing and the associated techniques involved in making-through-growing (as illustrated in Chapters 3 and 4). A systematic review of the literature and the analysis of various projects contributed to addressing these initial questions. As these questions were resolved, a new set of questions arose, needing further exploration (Figure 1). To review whether biological systems are parametric and, if so, what type of parametric system they represent, the study used RtD in Chapters 5 and 6. As in previously mentioned research projects, design experiments were conducted to test ideas and provide answers to design-related questions. Each answer derived from a design experiment generates additional questions, leading to a continuous and iterative process of exploration. After understanding the linearity and non-linearity of living materials' responses through experiments guided by the scientific method, the question of their predictability emerged. RtD approaches were once more used, with the help of visualisation methods, to explore a potential biofabrication method based on predictions in Chapter 7. The outcome of this investigation resulted in the proposal of a new design method for living systems, aiming to harness their predictability.

2. Methodology

2.1. Mixed Methods Methodology

In this research, a mixed-methods methodology is used to comprehensively explore the possibilities of working with living materials in design and fabrication. **Mixed methods** research involves researchers integrating aspects of both qualitative and quantitative research methodologies (Johnson, Onwuegbuzie, and Turner 2007). This includes incorporating qualitative and quantitative viewpoints, data collection, analysis, and inference techniques for a comprehensive and in-depth understanding, as well as to validate findings through the convergence of different research approaches (Johnson, Onwuegbuzie, and Turner 2007).

In this thesis, a blend of methodologies is applied to facilitate exploration through:

- Literature review to comprehend the current state of living materials' fabrication techniques and to position the chosen fabrication approach within the broader field.
- RtD to explore the possibilities of working with living materials in design and fabrication and to allow for an experimental approach to understanding the creative aspects of integrating living materials into the design process.

In addition to these methodologies, a combination of hybrid methods was employed:

- Lab-based successive design experiments to generate data and biomaterial probe methods,
- Quantitative and qualitative methods to analyse the design experiment outcomes, integrating the results within a scientific framework, ensuring a rigorous examination of both numerical and experimental aspects of the research findings,
- Ultimately, a method that was developed using visual representation, to provide an additional tool for data interpretation.

The combination of these diverse methods and methodologies enables a comprehensive understanding of the possibilities of working with living materials in design and fabrication.

2.2. Literature Review

A **literature review** is a systematic way of collecting and synthesising previous research to create a base for improving knowledge and elevating theory development (Snyder 2019). The review methodologies diverge according to the purpose, research question, search strategies, evaluation, and contribution of the research. In the case of this thesis, the purpose of the review was not to summarise every paper ever written, but instead to combine viewpoints to produce new models to design with the living. This approach is called an **integrative review** (Snyder 2019). It aims to critique the literature on bio-fabrication, allowing new theoretical frameworks and aspects to emerge. It is more than a simple description of the existing literature.

In this research, the literature review was conducted to gain a comprehensive understanding of the current state of living materials' fabrication techniques and to position the chosen fabrication approach within the field. It also helped in an in-depth understanding of existing generative architecture design methods (like parametricism) and their possible application to the fabrication of living materials[1]. By analysing existing literature, it was possible to establish common terminology, identify key trends, and capture varying perspectives within biodesign as a developing field. This process also facilitated the generating of a language for the proposed system. A literature review and evaluation of the terms and definitions[2] used in this area became essential for formulating new terminologies and effectively expressing novel approaches. The focus was on the design strategies adopted by practitioners, their role in the design process, and the definition of the final outcome. Ultimately, this methodology provides a foundation for further exploration and advancement in the field of biodesign.

2.3. Research Through Design

To explore the possibilities of working with living materials in design and fabrication, and to achieve the objectives outlined in Chapter 1, **design research** (which is an overarching approach) is followed. Glanville introduces the idea that design research is the scientific attitude of designing by perceiving a situation as a contextual challenge that requires understanding and a solution. He further suggests that it should be conducted through a systematic analysis of well-founded pieces of evidence (Glanville 2018). Consequently, rather than simply using the existing theories, this approach tries to create tacit knowledge by designing. In this case, tacit knowledge is not created by theory alone, instead, design research unites theory and practice. While using theory, an observer positions themselves outside the system to describe it, and in applied practice, the observer incorporates themselves actively into the system (Glanville 2018).

There are various design research approaches, with "**Research through Design**" (RtD) being one of them. RtD, used in this thesis, specifically focuses on using the design process as a method, aims to generate new knowledge and produce tangible outcomes. This meth-

[1] However, it was crucial to remember that when transferring concepts from one sector (digital architecture) to another (fabrication of living systems), there isn't always a direct correlation between the application and literature. Applications in the physical world may perform differently from the envisioned version. Therefore, additional measures like design-led research investigations were required.

[2] Key terms include biodesign, living materials, biological materials, tendencies, capacities, making, growing, biofabricating, bio-digital fabrication, linearity, correlation, plasticity, probabilities and so forth.

odology requires the active participation of an intentional observer in the design (Wolfgang 2018). The process starts by addressing and analysing the cause of a problem, moving on to setting goals to suggest solutions, and ending with the potential improvements of the problematic situation. In short, analysis, projection, and knowhow generation compose the RtD scheme (Wolfgang 2018).

The application of the RtD methodology within this research involved various methods. Specifically, to investigate the parametric fabrication of living materials and assess their potential as a biofabrication method, the research used probe methods, scientific methods, and visualization methods. Then the outcomes of these diverse methods were observed and analysed, forming the basis for proposing a method that addresses the objectives of the research. As a result, the selection of RtD was justified by its systematic approach, allowing for a comprehensive exploration of the living material design and fabrication process.

3. Methods

3.1. Biomaterial Probe Method

In order to test and evaluate the use of living materials, experimentation and hands-on engagement from the designer were required. Therefore, to evaluate the design outcomes, essential design actions such as prototyping, modelling, and experimenting were employed, leading to the recognition of the significance of creating prototypes and probes in the research process. **Prototypes** were used to gain experience, discuss future scenarios, and test theories to develop meaningful understanding (Stappers, Visser, and Keller 2018). They become ‘landmarks for reference’ in the process of a project (Stappers, Visser, and Keller 2018). Simultaneously, **a design probe** was used to investigate the matter and to inspire ideas. Probes helped to understand and test design suggestions to simulate the designer’s thoughts as a form of feedback (Ramirez-Figueroa 2017). Although both a probe and a prototype are used in the early stages of the design process, the difference between them is that prototypes get more refined over time. While prototypes are physical manifestations of ideas/investigation, probes remain a conceptually investigative tool and are materials designed to provoke or elicit responses (Sanders and Stappers 2014). Therefore, prototypes are employed to test products, while probes are used to test ideas.

After conducting a literature review to gain a foundational understanding of the field of biodesign, it was decided to employ the probe method to further explore and experiment with the subject matter. The concept of probes originated with the notion of ‘cultural probes,’ which were initially employed to reveal people’s values and activities towards a new idea. This approach has been extended to foster innovative systems for the investigation and integration of the physical and digital world (Gaver 2021). Gaver, Dunne, and Pacenti used cultural probes, which were objects, to gather responses and data about their specific design strategy. Subsequently, Carolina Ramirez adapted the probe method to biomaterials, using biomaterial probes as tools to directly engage with biological systems and generate new ideas and questions about designing with biological systems (Ramirez-Figueroa 2017b). She focused on the design process rather than the output by exploring the design possibilities of living materials through experiments with bacteria. As stated in Table 2, she used various design approaches, including parametric and diagrammatic, to evaluate how cells morph and assemble. The biomaterial probe method enabled the exploration of how biological systems can transform the nature of the design.

This research used the same method of biomaterial probes to assess the implications of biodesign and fabrication. Every intentional attempt towards that goal could be considered a **design experiment**. They rely on the premise that some features of the material can only be understood by interacting with it (Ramirez-Figueroa 2017b). Rather than illustrating data, they aim to provide a context to question how to design with living material engagement. They were used as the primary tool to gain new insights and information through the exploration of imagined scenarios.

These experiments were intentional attempts to achieve the research goals, and were carried out as part of the research through design methodology. By investigating imagined scenarios through experiments, it was aimed to deepen the understanding of the subject matter and inform the design process.

In this research, design experiments were flexible processes employed to generate new questions and speculate on new material systems in the field of biodesign. Since the outcomes of these experiments were uncertain, the questions formulated from the results of them could not be completely predetermined. As a result, adjustments to the design strategies might have been necessary based on the obtained results.

While experiments such as morphology studies conducted with mushrooms may not initially seem directly applicable to architecture, they hold conceptual significance by proposing a biofabrication framework. Morphology, alongside properties such as strength, flexibility, density, and other functional characteristics, is a defining feature of living materials and can be shaped through the material growth. By harnessing the agency of living materials as active fabrication agents within a parametric design framework, these experiments demonstrate the potential to influence not only form but also the functional characteristics of materials. This approach makes living materials applicable to real-world architectural contexts, where utility and performance are crucial considerations.

3.2. Scientific Method

Design experiments used rigorous scientific methods. However, the meaning of ‘experiment’, used previously in Section 3.1., might cause confusion. The design and science fields have different perceptions of what an experiment is (Ginsberg 2014). In design, an experiment commonly refers to a process that has no limits or end, and that has a goal of formulating new ideas; whereas, in the general principles of science, an experiment traditionally serves as a tool to produce data in order to test a hypothesis (Gauch 2003). Controlled experiments, used in scientific research, usually examine the impact of a planned modification on a certain quantifiable performance indicator (Anderson and Anderson 2019).

In this thesis, the experimental practice started with **preliminary tests**. Preliminary tests were an effective way to introduce a living material to the designer without a microbiology/ lab/science background. They proceeded through trial and error with scientific rigour while still creating space for ideas to be tested. They functioned as empirical laboratory work involving direct contact with the living material. Observing and exploring the organism's behaviours, life cycle, imitations, functionality, and properties through preliminary experiments provided valuable information that could be used to guide and inform the main science-led design experiments. They ensured that resources were used efficiently, and potential problems were detected in advance. Considering the decisions made from the preliminary tests, the main experiments were designed and conducted. Although the preliminary tests seemed to prolong the process, they helped to set the research on the right track by framing the scope of the research. Therefore, repeated preliminary tests were made to determine how the actual experiments could be prepared.

Tacit knowledge was cultivated through preliminary design experiments, which provided deep familiarity with the behaviour of the living organism beyond purely scientific or technical insights. This process facilitated a hybrid understanding, combining tacit knowledge (gained through hands-on experience and direct interaction) with explicit knowledge (grounded in biological principles), leading to an evolving comprehension of the material's capacities and responses.

The main experiments, building on the preliminary ones, aimed to test the viability of a concept or idea while striving to achieve a reproducible outcome. The focus was on understanding the behaviour of the organism, specifically in terms of its morphology, and how it can be influenced and guided. As such, controlled design experiments were devised and only one variable was changed at a time, with all other variables kept constant to explore the impact of the environment on fungal morphology. The desired physical and chemical environmental conditions in these experiments were created using digital tools. Digital tools, such as a growth chamber, provided standardisation and ensured that variables were accurate, reliable, and comparable between different trials when conducting the experiments. They were quantifiable, which helped in recording and plotting results, as they could be given a numerical value. This allowed for precise and objective measurements of the effects of the manipulated variable on the outcome.

The results of these experiments were assessed using mixed methods, including both quantitative and qualitative techniques. Subsequently, they were used to provide an iterative dataset that can inform future design decisions. Integrating mixed methods of analysis allowed for a balanced and comprehensive understanding of the results. The **quantitative methods** provided numerical data that allowed the analysis of patterns and relationships in the data. However, due to the absence of an established standardised method and tools available for this research, new quantitative measuring methods were developed. For example, digital files produced via 3D scanning records were precisely measured in terms of volumetric growth and size of different parts of the organism. The use of quantitative methods in this way provided a clear and objective way of analysing the results and enabled a deeper understanding of the physical properties of the living material.

On the other hand, **qualitative methods** provided a comprehensive grasp of the experiences of the researcher(s) and allowed for a more nuanced interpretation of the results. In cases where a satisfactory quantitative method was not available to describe certain morphological characteristics, such as texture, qualitative methods were used, such as microscopic imaging to capture and observe the smaller structures, details, and features. This helped to demonstrate how changing conditions affected the morphological characteristics of the organism and provided a visual representation of the data.

3.3. Visualisation Methods

The visual representation of the exploration of fungal systems was an important part of the mixed methods methodology. Photography, microscopic imaging, 3D scanning, and modelling were used to document the results of the experimental practice. This visual documentation provided a tangible element to the research and functioned as an act of recording the observations of the research-led designers.

In addition to simply recording the results, these graphical visualisations were used to define the results and contribute to the design process. They helped to drive the research in a specific direction and contributed to the strategic understanding of the results. They were used to seek a new relationship between form and context by targeted and methodical search (Harland 2018). In the shift from recording results to their representation, the graphical design contributed to shaping the results strategically. There were visual goals to achieve in graphical representations of the process. As Bolton suggests, visual thinking methods are a blended research method that fosters a design attitude through the adaptation of "designerly ways of thinking" in order to tackle complex and ill-defined problems (Bolton 2018). In this research, the graphic representation operated as a mediator to establish a dialogue between the designer and the matter, placing the practitioner in direct contact with the materials and tools.

Overall, the graphic representation of the experimental practice was a crucial component of this research, as it helped to bridge the gap between the abstract concepts and the tangible results of the experiment. It served as a visual thinking method that aimed to clarify ideas, demonstrate innovation opportunities, and assist other designers.

4. Research Materials

Fungi were chosen as the organism to be used as a biomaterial probe in the experiments for this research. They were a good fit for several reasons. They had the benefit of being complex, in terms of morphology and developmental plasticity, exhibiting significant phenotypic variation given the same genetic information. They could also be grown quickly. Producing a fungal product took, on average, two weeks, and fruiting bodies up to five weeks. This is a shorter time frame than plants, which can take months or years, giving fungi a clear advantage in time-limited research. However, not all fast-growing organisms were suitable to work

with. For instance, the common bean plant was a relatively fast-growing plant, capable of growing at an average rate of 3 cm/day, but it lacked developmental plasticity[4], offering limited forms (Wahby et al. 2018). Fungi, especially in the fruiting body form, possessed a high degree of morphological plasticity, enabling them to adapt to challenging conditions (Moore 2010). Consequently, they stood apart from other fast-growing organisms. In addition to the rapid growth rate and plasticity of fungi, fungal fruiting bodies have totipotency[5] that enables a single base cell to offer a wide spectrum of potential end products (Kaul and Ventikos 2015). Therefore, totipotency allowed for morphological variety in the growth process, presenting a broader scope for the designer's intervention.

In addition to structuring various morphologies in a short time from a single cell, fungi in the form of mycelium-based composites offer a vast array of other functions for the built environment such as building materials (Almpani-Lekka et al. 2021), furniture (Ross 2017) or packaging material (Holt et al. 2012). Furthermore, mycelium serves as an infrastructure for the transfer of materials across colonies, enabling the transport of materials, such as nanoparticles, over vast distances spanning hundreds of miles. Interestingly, feeding nanoparticles to fungi roots is employed not only for transportation but also for remodelling the network itself (Adamatzky et al. 2019). Living fungi can also establish a symbiotic relationship with other organisms. They can host organisms like algae by providing carbon and energy (Watkinson, Boddy, and Money 2016). This feature enables fungi to serve as a reinforcement or scaffold for the formation of other living materials.

In this research, I chose oyster mushrooms because of the availability of information, cost, proximity to the spawn supplier, and the simplicity of creating a physical environment for their growth. *Pleurotus Ostreatus* (Oyster mushrooms) were considered the most suitable strain for this research due to their prevalence, and non-toxicity to human health. Other commonly used fungus species in biodesign, such as *Ganoderma lucidum* (Reishi), *Ganoderma resinaceum* (Lingzhi), and *Trametes versicolor* (Turkey tail) possess distinct properties and qualities (Attias et al. 2020). Some of these strains exhibit a more wood-like quality and greater robustness in comparison to oyster mushrooms. Although oyster mushrooms do have drawbacks - their delicateness renders them susceptible to damage and leads to weaker structures. However, the focus of this research was not on structural stability and durability. Accessibility and ease of fungi production were of greater importance to the success of the work. Design experiments using oyster mushrooms as biomaterial probes did not necessarily need to result in the practical application of the living material or the production of a building material. Instead, the main objective was to demonstrate a broader concept of guiding living materials with inherent tendencies. Therefore, when evaluated against these criteria in comparison to other species, oyster mushrooms emerged as the most suitable strain for this research. As a result, they served as experimental operators to gain knowledge and to make these insights accessible for future investigations.

[4] *Plasticity is explained further in Chapter 5.*

[5] *Totipotency is explained further Chapter 6.*

5. Summary

The ultimate goal of this research was to create a vision for the future by testing the idea of designing with living materials, developing a novel technology, and experimenting with its compatibility with an organism. To do that, the methodology chapter has provided a comprehensive overview of the research framework and strategies employed to address the objectives of this study. The process used mixed methods and started with a literature review, as there was a need to deeply understand the various approaches to fabricating design with living systems. The literature review also helped to intertwine existing architectural design practices with the realm of living materials, as a method to inform them. As Salarian stated, taking inspiration from, and collaborating with various fields and philosophies enriches discourses (Salarian 2019). However, for this research, there was a need to validate the applicability of the parametric design approach as a biofabrication method. At this point, the synthesis of knowledge by design research functioned as a system to verify the design theories or ideas with real-life applications on living materials.

The use of the mixed methods methodology has facilitated the exploration of the design and fabrication of living materials in the design field. RtD allowed me to test ideas that emerged from the literature and to answer several sub-research questions. Throughout this research, each question naturally led to the subsequent one, and RtD, with the biomaterial probe method helped to answer them. Using fungi as a material probe helped to test ideas as proof of concepts and generated data. Data was produced as a result of design experiments which were run using mixed methods including scientific methods and visualisation techniques. Scientific methods allowed for a thorough examination of the experiments, research questions, and objectives. In order to show a connection between digital inputs and biological processes, visualisation methods were used. Various visualisation techniques not only presented the outcomes of the experiments, but also served as a method to predict the behaviour of the materials.

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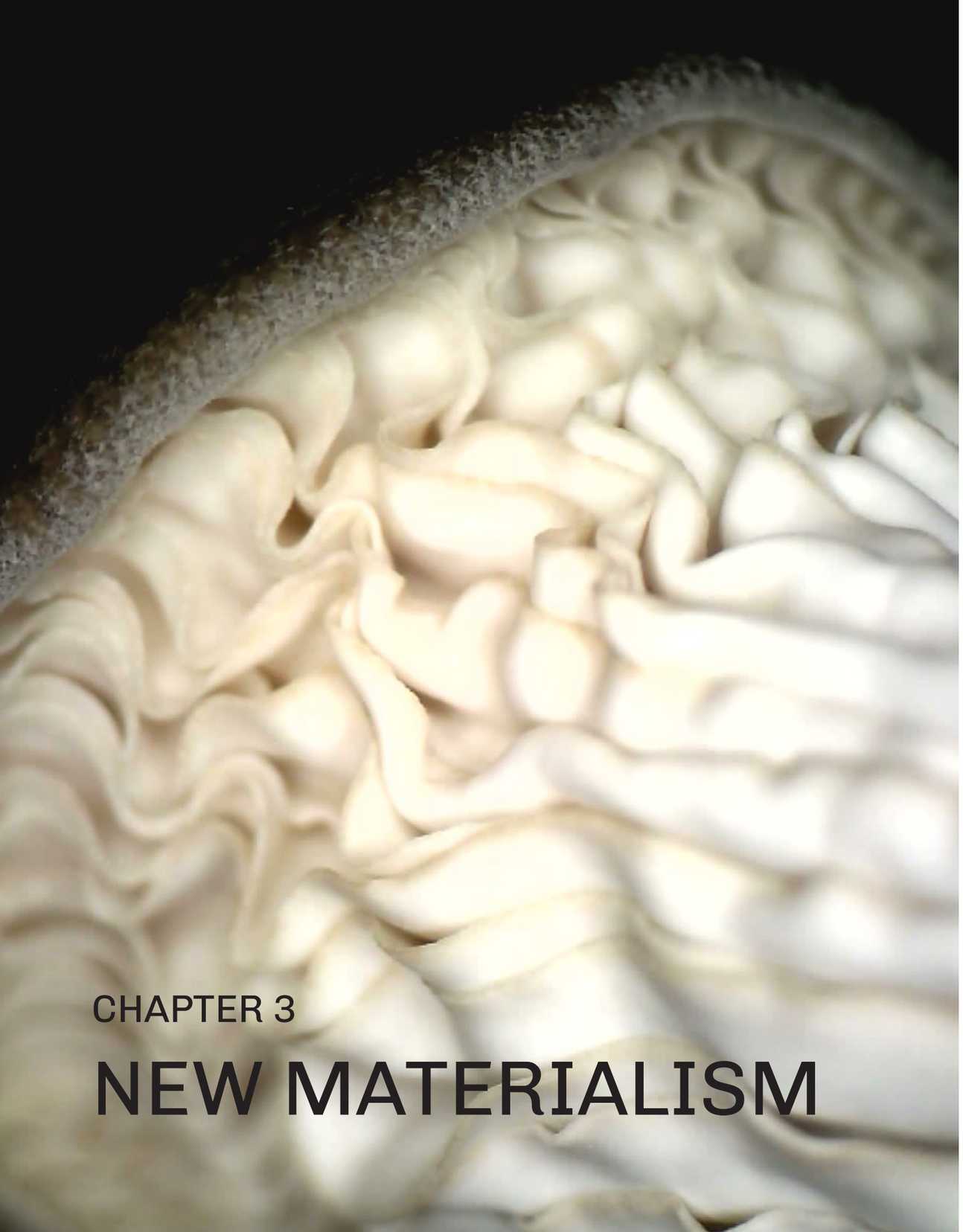

CHAPTER 3

NEW MATERIALISM

1. Introduction

In the biodesign field, there are different philosophical and ethical approaches to designing and fabricating using living systems. Going beyond biomimicry and using algae, fungi, bacteria, trees, or certain microorganisms as a material, requires nurturing from the designer. This care and support can also be considered a part of the biofabrication process. However, it causes ambivalent situations to arise, and the terminologies used, to be questioned. It becomes essential to define (i) what living materials are capable of doing and (ii) how they differ from normative materials commonly used in architecture. These distinctions are significant because they affect the way designers approach and interact with biological systems as their materials in their creative processes. Additionally, it helps to understand why and how it could not be ideal to treat biologically grown materials as if they have a low level of inertness.

After clarifying the distinction between making and growing, this chapter then conducts a systematic analysis of the literature review, focusing on various approaches to fabricating living materials. This understanding of the diverse approaches helps designers to select appropriate strategies and methodologies that align with their ethical stance and objectives. Furthermore, it highlights the potential for innovation by harnessing the inherent capabilities and tendencies of living materials.

A biodesign approach that acknowledges the material's ability to self-organise without external command aligns with a **New Materialist** perspective. Therefore, this chapter continues by exploring the applicability of a New Materialist approach to living systems. Living materials distinguish themselves from less inert materials, due to their capacity for self-assembly and developmental processes that lead to specific morphologies with unique exploitable features. Many current biofabrication approaches treat biologically grown materials as if they are similar to inert materials. Consequently, there is a need to develop tools that enable us to understand and work with these 'tendencies and capacities' while recognizing that we interact indirectly with the material through developmental parameters.

2. Making versus Growing

An **artefact** is defined as generated by art instead of nature (Simon 1969). Antiquities, tools, and art objects are some examples of artefacts, and they are turned into artefacts by being processed. The **act of making** transforms the material into an artefact (Hallam and Ingold 2016). Making occurs in a domain that is exclusively human, which is considerably different from plant and growing domains (Hallam and Ingold 2016). Although humans (designers or crafters) dominate the making process, that does not mean materials used while making have no natural tendencies or properties that can influence the process. Like all other entities, non-living things such as metal, plastic, concrete, and glass have agency. They have the capacity to act and reflect, as they have **material agency** (Knappett and Malafouris 2008). However, the sense of control over the reaction of the material has different levels (Gallagher 2007). Although non-living materials also possess complexity, they are on the lower end of the responsiveness spectrum compared to living materials. Therefore, while designing with non-living materials, designers or crafters would have more control and influence over the material within current expertise.

On the other hand, living materials are generated by organisms in nature and they grow. They are dynamic, self-assembled, and responsive to the environment they live in. Their behaviour can be unpredictable, and they would not always take the predetermined forms chosen by the designer. Fungus, algae, and plants are examples of materials that grow with or without care and nourishment. Human care and protection during the growth process help to use them for different purposes. Ensuring that the organism receives enough nutrients and water, eliminating competition, and even regulating environmental factors, such as temperature and light exposure, are examples of how humans provide protection and care through nurturing (Hallam and Ingold 2016). Intervening in the growth process of an organism to achieve desired outcomes leads to an ancient, yet deeply rooted, debate on the interplay between **nature and nurture** (Hallam and Ingold 2016). Natural growth is driven by inherent biological forces within the organism, whereas nurturing is influenced by external environmental factors.

From a distance, the distinction between **making an artefact** and **growing an organism** is clear. However, in biodesign, when we try to design with living entities by forming an organism with human intentions or skills, the clear distinction begins to blur. Making replaces growing and the living material turns into an artefact. The designer becomes the person who is **growing artefacts** or **making-through-growing**.

Making-through-growing represents a relatively new paradigm where living materials actively contribute to the making process through their inherent agency. These materials monitor and respond to their physical and chemical environment to achieve specific goals (Davies and Levin 2022). The designer participates in the process by altering the conditions under which the living material grows. By influencing factors like temperature, humidity, or nutrients, the designer encourages the material to grow in specific patterns.

Living materials that thrive on nourishment possess inherent factors and environmental conditions that influence their properties. However, does nurturing them with human intervention change their value (or essence) in terms of qualities or characteristics? Confusion may also arise as to whether a material is alive or not. Ingold explains this binary state of categorising materials through basket making (Ingold 2019). He uses basketry to illustrate his point, because the transformation of plant to material and from material to a finished object challenges our preconceptions of the distinction between artefacts and organisms. On one hand, each stage of basket making can be perceived as separate from the other. The willow can be viewed as a **living organism** whilst growing (or being cultivated), then when cut by humans it becomes a **material** upon its **death**. Willow, as a material, can be made into an **artefact** with human labour. Willow sculpting, where the living tree is guided into shape, shows a different perspective on the action of making a basket. Shaping the growth helps to ensure that the willow complies with the production process, allowing the human designer to treat it as a material despite the tree still being living. Therefore, this process blurs the lines of the living-to-material-to-artefact cycle previously described. As Ingold states, it is considered that any potential future “growth” that the plant may undergo would now be controlled by the nurturing power of human hands, which govern the process of turning raw materials into artefacts through the act of making (Hallam and Ingold 2016). Therefore, the boundaries between making and growing are becoming vague with human intervention in the growth process.

Working with fungi as a material also accommodates the intertwining relations of making and growing in various cases. Mycelium can be used when it is still alive [such as in the Bioknit (Scott et al. 2021) and The Living Room project (Scott et al. 2023)] in a dormant state or after it is dead [such as in the Hi-Fi Tower project (Benjamin 2014)]. If the material is to be handled in its living form, this means the process of producing mycelium is a growing activity, but guiding it through the growth process and using it as a material is an act of making. Making characterises an action in terms of its ability to produce a specific object (Hallam and Ingold 2016). Making an artefact with fungus, in a tailored manner, can only be achieved by feeding the organism; changing the environmental conditions (such as oxygen concentration, humidity, light intensity, and temperature) and allowing its intrinsic growth under these specific physical circumstances. Therefore, the distinction between making and growing becomes indistinct and the process becomes **making-through-growing**. To be able to control this process, skills are needed. Understanding and building up knowledge of an organism’s properties and nature will help the producer develop ideas and ways of use in the field. As engineers Vallas and Courard stated, using nature as it is alone cannot result in genuine “Living Architecture”; shaping nature to the deserved confirmation gives the true one (Vallas and Courard 2017). In short, the only way to craft a living material and, thus, turn it into an artefact is to collaborate with it while it is growing. In this way, methods of producing living architecture differ from the traditional ones. The dual status of being an organism and an artefact requires establishing a dialogue with the living. While growing a living artefact, the fabricator needs to follow the responses of the organism as a part of their growth process. Different types of growing approaches will be discussed further in the next section.

3. Methods to Fabricate Living Materials

Every community of professionals, such as craftspeople, scientists, farmers, and artisans have their unique approach to interacting with their materials, and this is also valid when working with living materials. For instance, craftspeople are able to recognise life in metal and eventually work more effectively with it because they are more motivated by a desire to discover what a metal can accomplish than by a need to understand what it is (Bennett 2010). The same applies to designers and their approach to materials. For instance, architect Louis Kahn's question "What does a brick want to be?" reflects the concept of material agency in the context of architectural design (Dade-Robertson 2021). Material agency implies that every material, including brick, has qualities, characteristics, and potentials that influence how it can be used or what it naturally wants to become. Despite the differences in materials, all professions try to interact with their material as active agents in the design and creation process, rather than passive elements.

The ethical considerations around working with living materials stem from different views and perspectives on nature and its role in our lives and the built environment. Throughout history, different cultures and ideologies have had different views on nature, ranging from romanticism's idealisation of nature to modernism's attempt to dominate and surpass it with technology (Modernism: Characteristics 2015). During the post-modernist era, there was a shift away from radical thoughts and a growing scepticism regarding the dominance of technology and this change prompted a more collaborative approach, bringing together nature, science, and technology (Leiss 1994). This evolving perspective towards a holistic and integrated view of nature holds the potential for a significant impact on the field of architecture. It could open up new possibilities for the integration of living materials into the built environment.

Agriculture is often considered the first successful attempt by humans to communicate with nature and cultivate organisms (Bookchin 1972). By controlling the conditions of growth and development, humans have been able to shape the form and behaviour of plants in specific ways (Ingold 2000). Concerning design, avoiding subjugating nature involves allowing the features of an organism's natural growth to develop by guiding it, rather than forcing it into shape. For instance, Ingold opposes the imposition of preconceived forms onto raw materials, advocating instead the exploration of their inherent potentials (Ingold 2013). He supports this perspective by drawing upon architect Lars Spuybroek's design philosophy. According to Spuybroek, a skilled gardener or chef has the ability to not only perceive the current state of things but also intuitively anticipate their future direction (Ingold 2013). This view emphasises the importance of respecting both natural phenomena and the resources that nature provides. By collaborating with nature, designers can create artefacts while respecting the delicate balance of the natural world.

There are several methods used in biodesign to fabricate living materials. Some of the most used methods include:

Figure 1
Works of Diana Scherer (left), Company New Heroes (centre), David Benjamin (right).

Figure 2
Works of Eric Klarenbeek (left), Petra Gruber and Barbara Imhof (centre), Joseph Dahmen (right).

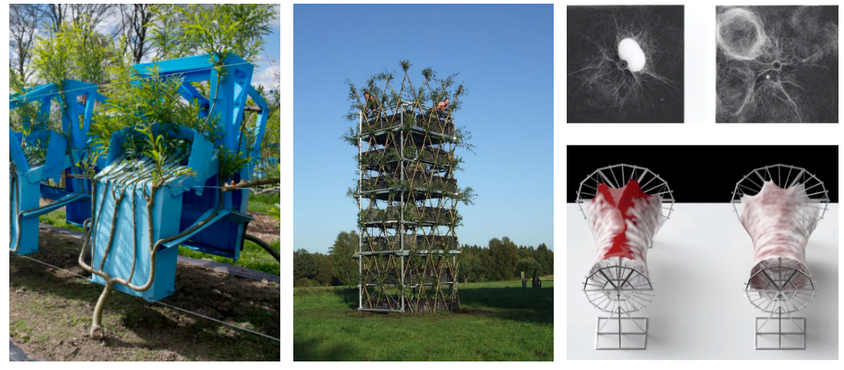

Figure 3
Works of Full Grown (left), Baubotanik Tower (centre), Silkworm Pavilion-II (right).

I. Changing the infrastructure that the organism grows in or on

The modernist approach conceptualises nature as a challenge to be overcome or something to be dominated by human capabilities (Leiss 2011). This approach aligns with the designer's top-down control in the fabrication processes. Diana Scherer, an artist who manipulates the root systems of oats and wheat to generate textile-like fibres through moulding, tries to reach clear-cut geometric patterns (Figure 1-left). She describes her work with self-reflection as follows;

> *"I send the roots in all directions. Maybe that's not very nice or respectful. If I wanted to respect the will of roots, I wouldn't make them grown in templates. I impose my will on them and domesticate them, but I'm not ashamed of that. Manipulation is the artist's challenge. I make nature do my bidding. And we all do that actually. Everyone prunes and mows, and we all want things to look exactly as we want them. That's manipulation too. What I do is no different really, it's just that you haven't seen this before." (Scherer and Kleis 2017)*

Cultivating mycelium in a mould is similarly an imposition and a constraint on the organism. 'The Growing Pavilion' by Company New Heroes (Company New Heroes 2019) and 'Hi-Fi Tower' by David Benjamin (Benjamin 2014) approach mycelium as a biodegradable bulk material rather than exploring its potential as a living organism (Figure 1-right). They see it primarily as a material that can be moulded into different shapes to create architectural structures rather than as a living system that can interact with its environment and evolve. By limiting the potential of mycelium in this way, they miss out on the unique qualities that come from working with a living material. However, the approach is still valuable as it contributes to the development of new construction materials and techniques that are sustainable and environmentally friendly.

In comparison to moulding, a less rigid way of using mycelium is semi-moulding, which allows, up to a certain point, collaboration with the material. Leaving openings at some locations on the mould helps to release the living organism, allowing it to act more freely. Eric Klarenbeek grows mycelium within windowed 3D-printed bioplastic substrates, and fruiting bodies appear at the openings (Fairs 2013) (Figure 2-left). Petra Gruber and Barbara Imhof cultivate mycelium in moulds made from various materials, shapes, and levels of openness. They use hard plastic, milled Styrofoam, and soft fabric moulds in different geometries, such as knitted basket-like moulds and grid shell 3D-printed scaffolds (Gruber and Imhof 2017). Dahmen and AFJD Studio try to demonstrate architectural softness through their 'Mycelium Mock-up' installation. They aim to explore material softness in line with manufacturing conditions and exchange with materials' surroundings (Dahmen 2017). In the installation, although they used traditional moulding, leaving the mycelium composite bricks alive[1] caused a response of the bricks to the environmental changes, helping them to liberate the material. Not surprisingly, after a while, with a temperature drop, the blocks started to fruit mushrooms (Figure 2-right).

In conclusion, from moulding to semi-moulding, there are two ways to interact with living materials. This range offers a new way of thinking about material use and production in architecture and design.

[1] If the fungi-based products are not exposed to excessive heat, they will continue to stay alive and show metabolic reactions.

II. Changing the cell's physical and chemical environment

Another way of interacting with the organism is where the final living artefact forms through an ongoing dialogue with the designer. In this process, the designer dynamically adjusts the context in response to the growth of the organism. This interaction can be described as a dialogue, wherein the designer establishes the conditions for the organism's development, and the organism, in turn, shapes its final structure and form in response. Designer and organism interaction results in a final product that is unique and shaped by both parties. In a way, it is a collaborative effort between the designer and the living material. This approach to design and fabrication emphasises the importance of respecting nature and its resources and not dominating them. It represents a shift from the modernist approach, which saw nature as a challenge to be overcome, to a more postmodernist perspective (Leiss 2011), which seeks to collaborate with nature and understand its inherent properties.

In applications like bonsai making and willow sculpting (by Full Grown Ltd, UK) designers are supporters of growth by nurturing, pruning, and inosculating[2] the living material (Figure 3). As a technique for plant-based shaping, the Baubotanik (Living Plant Constructions) group aimed to establish a theoretical and scientific foundation for living architecture, addressing a previous lack of such knowledge (Oommen 2015). They investigated and harnessed its aesthetic, constructive, and ecological potential within architectural design, landscape architecture, and urban planning (Ludwig 2014). Their primary objective was to shape and optimise the growth of trees for various purposes. Apart from pairing and interlacing branches, the group crafted their structure by using the ability of plants to replace pruned pieces. Encouraging new growth through pruning is crucial, offering opportunities to integrate foreign materials like metal and plastic (Oommen 2015). Another project that centred on shaping an organism through intervention in the physical and chemical environment is the Silkworm Pavilion-II project by designer Neri Oxman et al. They guided silkworm distribution across a woven surface by changing the direction, duration and intensity of heat and light. Additionally, they used a custom threading tool to control the influence of gravity on the worms (Oxman 2021). This control allowed them to ensure the homogeneous spread of fibres, aligning with their objectives since environmental conditions directly influenced silk production (Figure 3-right).

The common ground of these projects is their support for the autonomy of living materials. By nurturing the organism and changing its environment in a controlled way, the designer is able to guide the growth and shape of the final product, while still allowing the material to respond and adapt to its environment in its own way. This approach highlights the potential for collaboration and mutual benefit between humans and nature in the creation of new materials and designs.

[2] Pairing branches

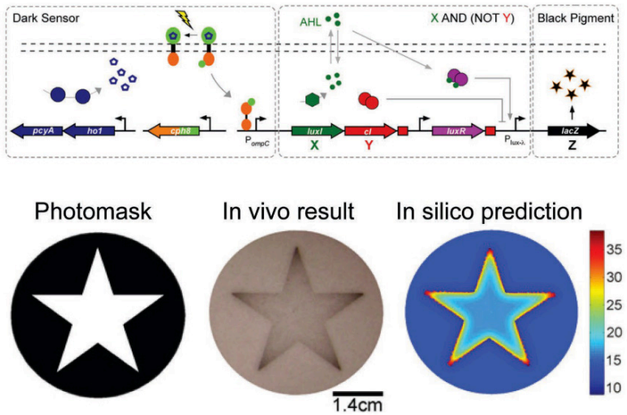

Figure 4

The experiment's photomask and a photograph of a bacterial lawn showing the edge detector circuit.

III. Changing the cell itself

This category includes engineering the DNA (or blueprint) of a living material to make it more resistant or to make it specific compounds of interest. Additionally, these modifications can add new genes to cells that help to develop materials with various mechanical or biochemical functionalities (Nguyen et al. 2018). For example, an edge-detecting organism was created (using genetic engineering tools) by integrating a genetic dark sensor that only activates cells that are physically next to the dark zones (edges) (Figure 4) (Nguyen et al. 2018). This integration of genetic elements creates a specialised functionality within cells, allowing them to respond specifically to environmental stimuli and produce specific outcomes.

In summary, when designing with living materials, there are primarily three approaches to fabricating them: by modifying the infrastructure in which the organism grows, by altering the physical and chemical environment of the cells, or by directly modifying the cells themselves. In the first approach, the designer defines the final form of the material in advance by direct physical intervention using tools such as moulding, 3D printing and CNC milling. Therefore, it can be referred to as a top-down fabrication method; the size, geometry, and openings of the mould or scaffold are predefined. On the other hand, the second and third approaches are less physically controlled or, at the very least, aspiration to minimize control. In these methods, living materials form larger structures through the development of new emerging relationships (Tiruvannamalai-Annamalai, Armant, and Matthew 2014). These two methods, involving the alteration of the growth environment and the modification of the cells themselves, can be categorised as a **bottom-up** fabrication approach. Zhang (2023) argues that growing one cell at a time without a predetermined form constitutes a bottom-up fabrication process.

The top-down approach may constrain some living materials' abilities within a predefined structure. The living material only fills the negative space in a confined setting, especially in moulding. Besides the technical challenges of using traditional top-down techniques, such as diffusion limitations, low cell density and non-uniform cell distribution (Tiruvannamalai-Annamalai, Armant, and Matthew 2014), defining the final product at the beginning of the process is a subject open for discourse. However, it should be remembered that the top-down fabrication approach is not a restrictive method for every living material, as some organisms cannot be fabricated without a mould or a surface to cling to. Organisms such as algae, urease-producing bacteria, and bacterial cellulose will always require a container to grow in because they need liquid nutrients, while fungi (in mushroom form) and some plants can self-stand and do not require a supporting structure to grow.

In the bottom-up approach, the material goes beyond the role of simply shape-filling (Oxman 2010b) and consequently, a new layer of relations starts to emerge. While making-through-growing, cells self-assemble into larger structures, and the form comes from the system as a whole rather than from individual components (Kretzer 2017). This allows material emergence. **Emergence**, in systems theory, means that the overall characteristics of larger systems, usually cannot be reduced to the properties of its constituent smaller parts and these irreducible properties are called **emergent** (Heylighen 2000). **Material emergence** refers to the ability of a material or system to generate new qualities or behaviours that are not explicitly specified by the designer or programmer (Ratter 2012). The restriction from the direct physical constraints, as in top-down approaches, reduces the room for material emergence. The more the designer constraints an organism, the more they reduce the quality/chance of emergence that living materials have, as organisms lose their inherent potential when cultivated in a predefined form (DeLanda 2005). Philosopher Manuel DeLanda describes it as the **degree of freedom** to state any relevant means of changes in the system (DeLanda 2007). Bottom-up approaches increase the degrees of freedom in living materials, as they allow material emergence. With higher degrees of freedom, the material has room to respond, adapt, self-repair, regenerate, and develop in various ways based on its environment.

In the case of living materials, the inherent potential or qualities of the organism can be influenced by the growth conditions and environment in which it is cultivated (as in Group II: Changing the cell's physical and chemical environment). If the material is cultivated in a highly controlled and constrained environment such as 3D printing or moulding (Figure 5 x-axis) (as in Group I: Changing the infrastructure that the organism grows in or on), with specific parameters set by the designer, the material would not perform some of its inherent potential, and would be less able to generate new qualities or behaviours (Figure 5 y-axis). On the other hand, if it is allowed to grow and develop in a more open and less physically constrained environment such as a tree growing in nature, it may have a greater ability to generate new qualities that are not explicitly specified by the designer. As a result, biofabrication processes involving minimal physical intervention facilitate the emergence of novel, coherent structures, patterns, and properties during the process of self-organisation in complex systems (Corning 2002).

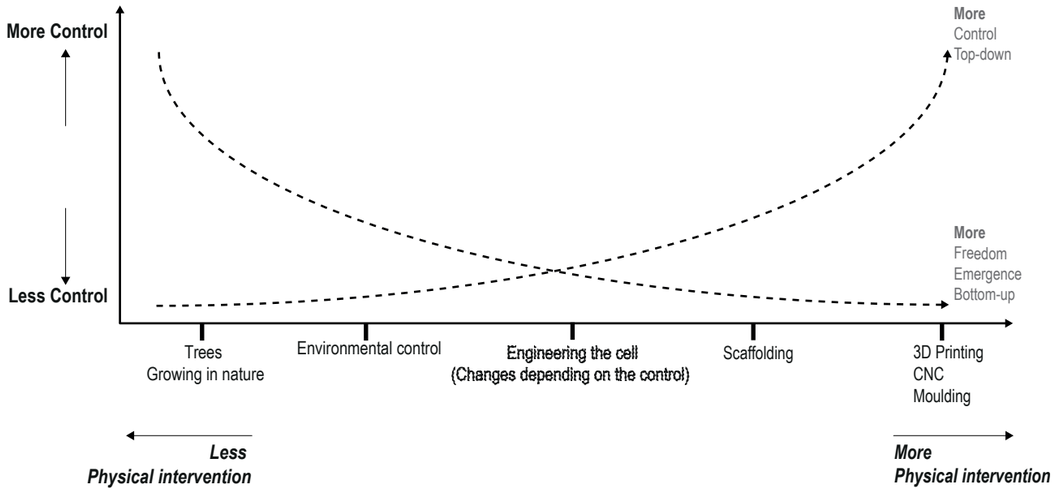

Figure 5

Diagram representing the spectrum of physical intervention in biofabrication processes (x-axis) and its impact on design approaches for material emergence (y-axis).

Another way to frame top-down versus bottom-up biofabrication processes is by referring to them as the "**design of living materials**" and "**designing with living materials**" approaches. Designing with living materials involves collaborating with cultured organisms and allowing them to behave as active matter. Even though human designers can manipulate cells, these cells can still influence the design process as "agential matters", exhibiting decision-making and problem-solving abilities (Davies and Levin 2022). In these complex processes, the designer's role is to shape the probability of achieving specific outcomes (within the limits of the living material's potential), such as a desired morphology, functional gradients, material performances or properties. This involves planning and predicting the material's characteristics or forms by creating and managing the conditions necessary for their development.

A critical aspect of this method is regulating the growth environment, often with the aid of digital tools, to guide the material toward the intended outcomes. This approach is relevant to architecture as it enables the creation of materials with tailored forms, functional capacities, properties and gradients. Aligned with the New Materialist perspective, it acknowledges the tendencies and capacities of materials, allowing them to influence their own development. As a result, living cells become both the object and subject of the design process, simultaneously acting as the "maker" or "fabricating agent", the "materials" and the "artefact".

The material, as both the fabricating agent and the material itself, is relevant to architecture because it allows for the creation of materials with tailored forms, functional capacities, properties, and gradients. Its plasticity enables the creation of structures or artefacts with properties fine-tuned for specific functions, adapting to the designer's guidance to produce a wide variety of material forms. This adaptability facilitates the "making with" biology approach in design.

4. New Materialism

The concept of New Materialism is applicable in the fabrication of living materials by allowing them to grow with minimal physical intervention. "**New Materialism**", the late postmodernism phenomena in architecture, is a movement that relies on cutting-edge material sciences, and the combination of engineering with design, and revolutionises the role of materials in architecture (Oxman 2010a). This movement is based on the notion that matter has the ability to generate form on its own and does not require external command (Manuel 2017). The New Materialist approach allows for the creation of materials and structures with greater degrees of freedom and for maximising the emergence of material properties.

In the field of biodesign, New Materialism has been particularly influential. Integrating living materials into design aims to create dynamic systems that are alive (as active matter that makes decisions and solves problems) and encoded in wetware, in addition to hardware and software (Dade-Robertson, Ramirez-Figueroa, and Hernan 2017). These systems behave as **active matter** empowered by their own **tendencies and capacities** (Menges 2015). This perspective on the tendencies and capacities of living materials is significant for biodesign, as it moves away from a human-centred approach to design, recognising that we

(designers) are just one part of a larger ecosystem (Bennett 2010). Even though human designers can manipulate cells (as in Group III mentioned above), those cells can still be considered as active matter, as Dade Robertson et al stated, due to their decision-making and problem-solving abilities (Dade-Robertson, Levin, and Davies 2023). By acknowledging the importance of materials and their inherent properties, the New Materialist approach has the potential to transform how we design and interact with the world around us.

To establish a framework for active materiality within the concept of New Materialism, DeLanda adopts a philosophical perspective and introduces five key elements. He names these loops, causality, morphological states, material properties, and capacities to conceptualise and bring a new vision to materiality (DeLanda 2015). **Loops** arise from a complex series of events and are necessary for maintaining homeostasis[3] (DeLanda 2015). Loops are metabolic cycles that help to regulate internal stability, enabling the material to adapt to a changing environment. For example, a living cell responds to environmental changes through enzymatic activities and adjusts its metabolic pathways. Given the cell's role as a material factory of biopolymeric building blocks, these metabolic cycles function to modulate the end product or the artefact (Nguyen et al. 2018).

[3] Any self-regulating mechanism that allows biological systems to remain stable while adapting to circumstances that are best for survival is known as homeostasis

Causality describes the cause-and-effect relationship between stimuli and changes in the material (DeLanda 2015). This relationship can be non-linear, meaning that the same input does not always result in the same output or the changes in inputs and outputs are not proportional (DeLanda 2015). External factors, including the physical and chemical environment, can alter the metabolic cycles of materials and cause chemical reactions within them. However, these metabolic cycles can be "hacked" by changing the molecule or cell itself or its environment through engineering tools. Even though a relationship can be established between inputs, such as increased nutrition leading to growth in living materials, this relationship is not always predictable. The complexity and autonomy of the material, resulting from the cycles, reduces the control that the designer has over the material. As a result, in the case of living materials, they become a non-human agent in the design process.

The states of a material describe its morphological responses to environmental changes or interventions. For example, the metabolic cycle of the living material adapts to new conditions, resulting in morphogenesis, the process of self-organisation, which gives rise to three-dimensional forms (Cogdell 2019). The morphological states such as size, shape, and position can change due to various stimuli (DeLanda 2015). Although active matter, such as living materials, have autonomous metabolic cycles and non-linear causality, it is still possible to estimate their response within a certain range.

The properties and capacities of materials exhibit remarkable versatility (DeLanda 2015). Properties represent quantifiable and measurable characteristics, such as form and material properties, under specific conditions. In contrast, capacities define what a material can do or how it responds to varying conditions. While material properties are static representations, capacities are dynamic and adaptable (DeLanda 2015). Living materials demonstrate diverse capabilities in various states, providing a wide range of possibilities for designers to

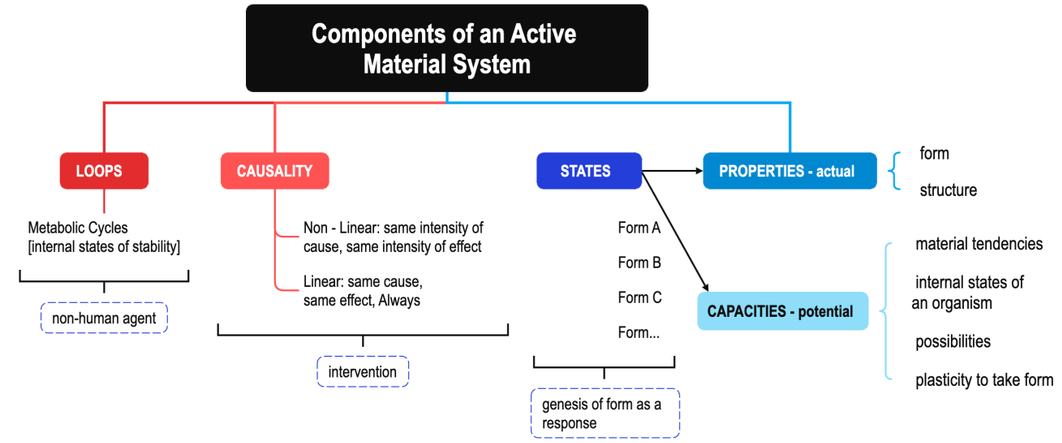

Figure 6

The framework of an active material system.

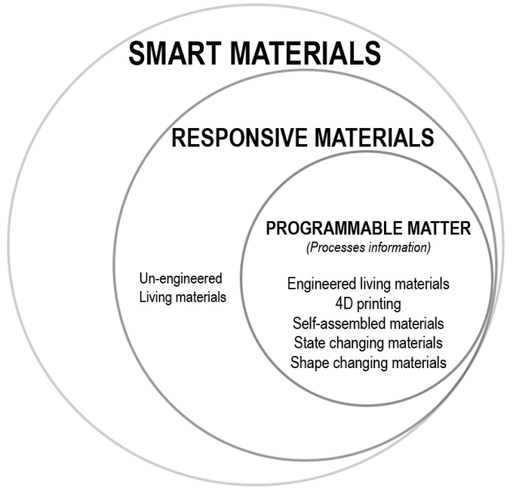

Figure 7

Diagram of clusters of smart materials.

explore. This versatility not only fosters creativity, but also leads to new, unexpected discoveries. For instance, consider fungi as a material; they can serve as a decomposer (O'Leary et al. 2019), binder (Saez et al. 2022), sensor (Adamatzky, Gandia, and Chiolerio 2021), messenger (Sherry, Dell'Agnese, and Scott 2023), bulk material (Elsacker et al. 2021), insulation material (Schritt, Vidi, and Pleissner 2021), furniture component (Ross 2017), and even a work of art (Sydor et al. 2022). This multifaceted utility highlights the diverse potential inherent in a single material. DeLanda recognises the significance of material properties and capacities as essential inputs within the framework of the New Materialist approach, referring to them as "immanent potentials" within a material (DeLanda 2015).

Figure 6 attempts to present the components of an active material system. However, it is important to note that this framework cannot be algorithmically represented for all active materials including living materials used in biodesign. There can be a range of inputs and outputs that are measured, but these are not exhaustive. Additionally, there are imperceptible external factors that may appear negligible but can have a significant impact on the system.

Materials with tendencies and capacities, which are active participants in the genesis of the form (Kretzer 2017), have given rise to the concept of smart, functional, multifunctional, and intelligent materials (Figure 7). These terms are often used interchangeably, but they refer to materials that possess inherent properties enabling the creation of structures or systems with "smart" behaviour (Kretzer 2017). **Programmable materials** can change their physical characteristics in a direct and controlled manner, such as shape, density, conductivity, and opacity (Kretzer 2017). **Responsive materials** and material technologies can respond to external stimuli, such as temperature, light, or pressure, and change their physical or chemical properties accordingly. They have the capacity to self-assemble autonomously and form patterns through the interaction of their constituent parts, exhibiting transformative and responsive characteristics. According to Oxman, natural forms are intrinsically sustainable since they are more effective than any of mankind's own material techniques and are directly informed by the materials from which they are made (Oxman 2010b). **Smart materials**, in general, offer complex behaviours and functionalities that traditional manufacturing methods may find difficult to achieve. Consequently, both living biomaterials and naturally occurring materials can be regarded as smart. The advancement of these materials opens up new possibilities for material design and application.

Living materials, being inherently smart and possessing a higher degree of material agency, can form on their own without being constrained by predetermined qualities imposed upon them. According to the New Materialist approach, they are considered as integral parts of human life rather than just raw materials (Corcoran, Moore, and Jazvac 2014), as they can be born, die (Tsaknaki et al. 2021), and actively get involved in design (Karana, Barati, and Giaccardi 2020). Therefore, as mentioned in the New Materialist approach, the way designers fabricate them should allow them to present their capacities, since they are relatively less inert materials; meaning, the biofabrication methods should allow material emergence, have a high degree of freedom, and allow the material to perform its tendencies and capacities.

5. Conclusion

This chapter delves into the distinctions between making versus growing, nurture versus nature, artefact versus organism, and various approaches used in the fabrication of living materials. The term "making-through-growing" was introduced to describe the artefacts that emerged and were nurtured as a biofabrication approach. After clarifying the terms, the chapter explored the theoretical understanding of living systems and the different approaches used to fabricate them. The biofabrication methods were classified based on the designer's intervention by (1) altering the infrastructure that the living material grows in or on, (2) altering the living material's environment, and (3) altering the cell itself. Within these fabrication methods, the concepts of top-down and bottom-up fabrication, material agency and emergence, were examined. It was concluded that excessive direct physical control over the living material reduces its emergence. Limiting the living material's degrees of freedom may potentially impede its ability to perform its unique capabilities, including growth, self-assembly, and morphological development.

As living materials are highly transformational during all stages of their lifetime, it is necessary to consider their inherent temporality as a core concern in architectural design concepts and tools to design with (Ramsgaard Thomsen and Tamke 2022). Therefore, the New Materialist philosophy which emphasises the ability of materials to generate their own form and function, was introduced. In DeLanda's framework for active materiality within New Materialism, four key elements were introduced. These include (i) Loops, which are crucial for maintaining a material's internal stability and responding to environmental changes; (ii) Causality, which explores the cause-and-effect relationship within materials; (iii) Morphological state, which represents how materials adapt and self-organise in response to stimuli; and (iv) Material properties, which are quantifiable characteristics, while capacities define a material's dynamic responses to changing conditions. These key concepts formed the basis for new methods that are applicable for making-through-growth and developing tools that enable designers to work with biological tendencies and capacities. By understanding this explicit material logic, designers can create more efficient biodesign technologies.

The chapter concludes by questioning the methods applicable to "making-through-growing" and the potential use of a New Materialist approach to engage with the tendencies and capacities of living materials. The next chapter delves into existing design and fabrication approaches from architecture that can be integrated into living systems, intending to explore novel biofabrication methods. This investigation seeks to answer how designers can interact with materials through their developmental parameters, while recognising that living materials have an element of unpredictability and instability due to their adaptability.

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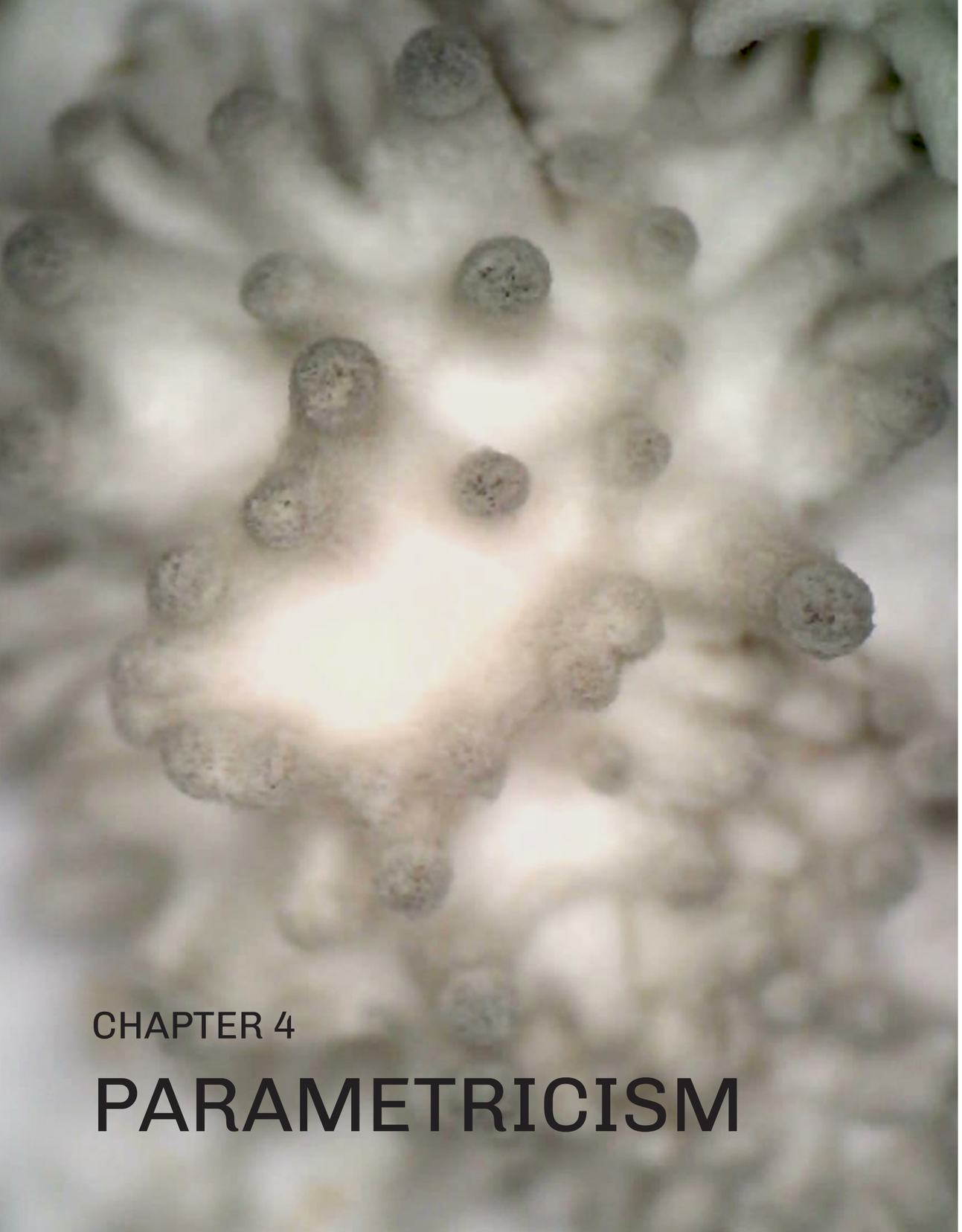


CHAPTER 4

PARAMETRICISM

1. Introduction

As discussed in the previous chapter, it is important to acknowledge that employing top-down fabrication methods, involving direct physical intervention to shape self-structured living materials, can restrict their material emergence, degree of freedom, and capacities (or potentials). Therefore, developing tools which allow us to understand and work with living systems' tendencies and capacities is crucial. Digital architectural design provides strategies that can be applied for both designing and fabricating living materials through-growth that allows interaction by environmental parameters. This interaction can subsequently lead to particular developmental pathways. A **developmental pathway** refers to the sequence of steps or stages that guide the development of an organism from its initial form to its mature state (Davies, 2023).

According to architect and researcher Rivka Oxman, there are different types of digital design methods used in architecture (Oxman 2006). Among these methods, **formation models**[1], which are digital processes that build an associative relationship to generate form, seem relevant to explore within this research (Kotnik 2010). Formation models allow the establishment of a direct or indirect relationship between form and designer. Interacting with the environmental parameters affecting the morphology of living material through its developmental pathway, is similar to generating digital forms using formation models, in terms of the designer's role and associative design process. In both cases, the act of making or growing relies on the manipulation of various parameters that influence the resulting form. Although comparing digital and biological systems seems initially tenuous, the design process involves setting (physical, chemical, and digital) environments as inputs that subsequently lead to distinct material properties, forms, and functionalities as outputs. These shared principles and practices between digital and biological design processes can offer valuable insights and inspiration for enhancing and advancing the field of biological making-through-growth.

[1] Explained further in Section 2.

This chapter questions whether a formal parametric relationship between biological fabrication and environmental conditions is feasible and, if so, what it might entail. To do that, it systematically analyses the processes of digital form generation and the morphogenesis of biological organisms from literature. It then examines various examples where biological artefacts are designed and grown using a parametric approach. These examples serve as the basis for a comparative analysis of the similarities and distinctions between biological and digital design and formation processes. Understanding whether a digital formation model is suited to designing with living materials helps to develop a biofabrication method that (1) applies to making-through-growing, (2) enables material interaction through its developmental parameters, and (3) allows designers to work with the tendencies and capacities of living materials.

2. Digital Form Generation

Form generation through digital processes, regardless of their complexity, relies on interaction and reflection with the designer (Oxman 2006). This interaction is crucial because in these processes, “the computer is not a neutral tool, but rather is actively shaping the way designers are approaching the question of design” (Kotnik 2010). Rivka Oxman investigates this interaction and introduces a framework for digital design models. Oxman has grouped digital design models into five classes to organise emerging design theories and methodologies from the early 1990s. These classifications depend on different relationships between the designer, the conceptual content, the design processes used, and the design object itself: Computer-Aided Design (CAD) models, formation models, generative models, performance models, and integrated compound models (Kotnik 2010). Based on Rivka Oxman’s schema in the **digital formation models**, form is generated by interacting with a digital method, such as the use of software that enables it. The methods are like scripting to interact with and work within a form generative environment that operates using **non-deterministic logic** (Oxman 2006). This means that the forms are not entirely predictable and can vary, unlike traditional CAD or paper-based design methods that produce specific, fixed representations of forms (Oxman 2006). Within this variability, this process is different from the traditional approach used in CAD, where designers work with explicit representational structures (Oxman 2006). The new type of designer engages with, manages, and modifies generative and performative mechanisms, essentially acting as a tool builder within the digital design process. The designer moves beyond a **form-maker** and creates a system that allows **formation** (Oxman 2006). The designer interacts with a digital framework that has been created by a mechanism through a set of predetermined rules or relationships (Oxman 2006).

Oxman groups the digital formation models into three subclasses: (1) **topological design**, which uses topology and non-Euclidean geometry to create design media for formation; (2) **associative design**, which relies on principles of parametric design and generative components; and (3) **dynamic design**, which utilises animation, morphing, and other motion-based and time-based modelling techniques to generate multiple instantiations within a dynamic continuum (summarised in Table 1 and Figure 1) (Oxman 2006). In digital formation processes, the (digital) form is established by relying on input and outputs or parametric relations. Design tools, such as CATIA, Rhino with Grasshopper, Maya, and Cinema 4D, use algorithms and mathematical equations to generate designs based on a set of inputs. By manipulating these inputs, designers can easily create variations and manage transformations while maintaining the topological relationship between elements (Oxman 2006). This enables the production of geometries, allowing greater flexibility in the design process.

The distinction between these design models lies in how the parameters influence the final form, whether through direct or indirect means. In topological design, input variables are directly linked to the final form, whereas in associative and dynamic design[2] relationships, there is a varying degree of indirect linkage, primarily due to differences in the underlying algorithms (Figure 2). However, despite these differences, all models prioritise the formation of iterative or adaptive forms over static and predetermined ones. As Kotnik stated, they rely on changing a set of parameters within a fixed field of geometric connections for interaction and control. Thus, all digital formation models are parametrically computable digital design techniques (Kotnik 2010). Consequently, the significance of parametric design becomes more pronounced. In this thesis, the term “**parametric**” is used to define correlations or relationships, such as associative or dynamic ones, to create designs and generate forms that respond to changes systematically.

In architecture, parametric design is an approach that uses a set of rules, algorithms, and variables to generate design options to explore multiple design possibilities (Gu, Yu, and Behbahani 2018). It is a process that allows designers to create and manipulate complex forms and structures using a set of parameters.

> *“…In parametric design, relationships between objects are explicitly described, establishing interdependencies between the various objects. Variations, once generated, can be easily transformed, and manipulated by activating these attributes. Different value assignments can generate multiple variations while maintaining conditions of the topological relationship.” (Oxman 2006)*

The role of the designer in a parametric design process is not to design the form of the object or system directly, but rather to define the key controlling parameters and their relationships (Jabi 2013). The design process is generally based on generating variables and relating these variables (to solutions) to produce variations, overcome complexities, and simplify and solve problems in terms of geometry and material compositions (Thomsen 2022). Therefore, the parametric approach requires working with **algorithms**, which create systems based on processes aiming to serve a goal using several steps (Dino 2012).

[2] The digital form is dynamically generated based on input variables and their impact on predefined material behaviour. In animation software, such as Maya or Cinema 4D, these behaviours can encompass material types (e.g., silk, concrete or metal, defining the strength of the vertex connections) and forces applied to the material (e.g., turbulence, gravity, wind, drag or vortex). The designer then sets a timeframe to capture the final form.

Table 1

A summary of the digital formation models proposed by Rivka Oxman.

	Definition	Design Outputs	Design Terms
Topological Design (Figure 1.1)	The study of the relational structure of objects rather than of geometry.	Morphology of complexities	Lofting, digital modifiers, hyper-surfaces, NURBS (non-uniform rational basis spline), blob architecture
Associative Design (Figure 1.2)	The use of parametric design techniques that exploit associative geometry	Interactivity, transformability and parametrically controlled variations	Variations, alternatives, parametric, designer as a tool builder
Dynamic Design (Figure 1.3)	The denial of classical notions of representational conventions such as static space	Interactive, dynamic, and responsive designs	Motion-based model, key-frame-animation, forward-inverse kinematics, dynamic force fields

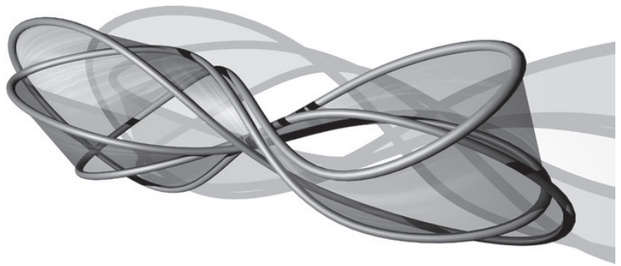


Figure 1.1

Topological Design

In the Mobius House by Ben Van Berkel, all the spaces are created through a single surface. Hyper-continuity of surfaces acts as programmable parameters to inform its design concept and offers flexibility by accommodating variations in design (Oxman 2008).

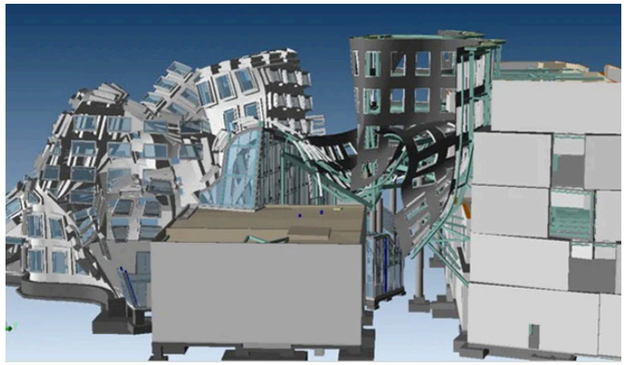


Figure 1.2

Associative Design

In Gehry Technologies's work, the Catia Model is used to create and manipulate non-standard geometry. Using associative design models, users can interact with integrated inner tools through scripting methods and parametrically controlled tools, enabling the generation of discrete structural variations during design formation processes (Oxman 2006).

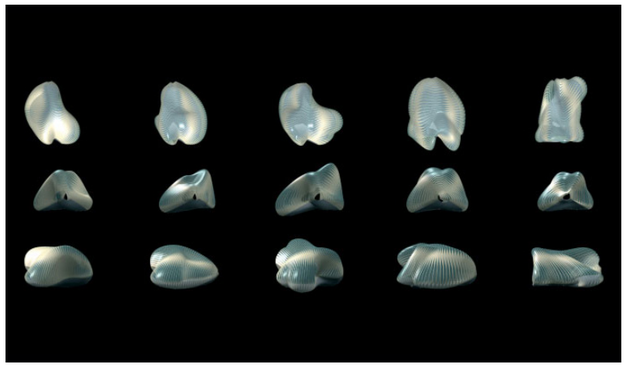


Figure 1.3

Dynamic Design

In Greg Lynn's Embryological House, design interaction occurs within the interactive framework of animation media, which generates the form, instead of the representational medium of the emerging form (Oxman 2006).

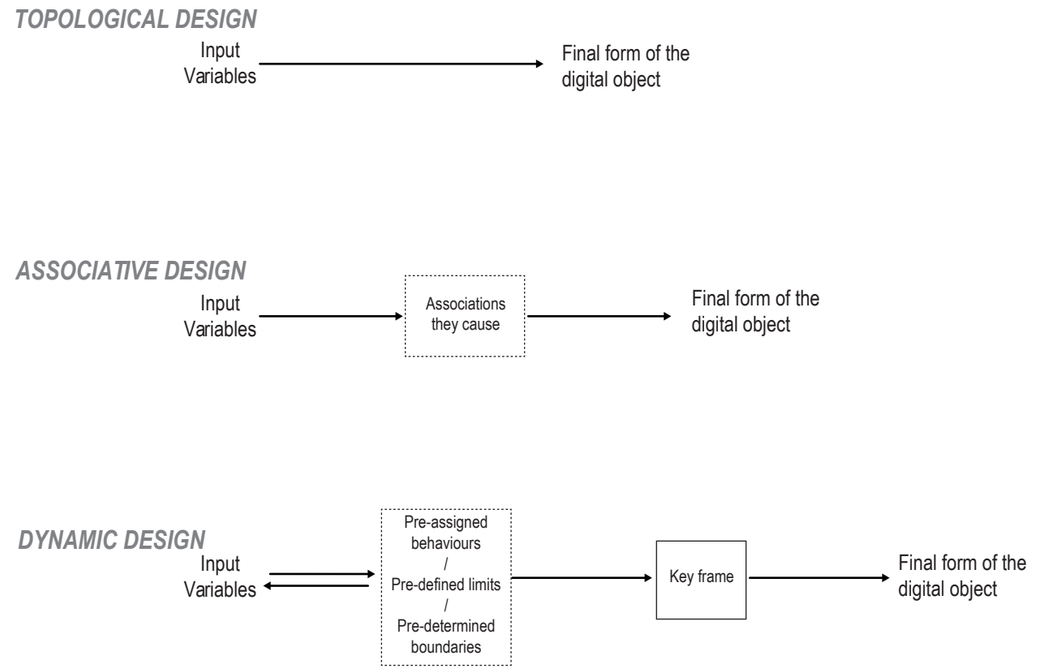


Figure 2

Diagram illustrating three distinct design models and how input variables affect the final form. Topological design directly links input variables, while associative and dynamic designs exhibit varying degrees of indirect linkage due to algorithm differences. In associative design, input variables create associations influencing the final form, while dynamic design generates form based on pre-assigned characteristics and timeframe.

"…An algorithm takes one value or set of values as **input***, executes a series of computational steps that transforms the input, and finally produces one value or a set of values as* **output***" (Dino 2012).*

Working with inputs that can be changed constantly helps to generate dynamic outputs. As a result, the output that a designer produces becomes "a generic object or an open-ended mathematical notation designed for interaction and variability" (Carpo 2017). This correlational relation of inputs and outputs is defined as an objectile by Bernard Cache and Gilles Deleuze (Carpo 2017). The **objectile** is restricted with specific parameters that make it a never-finished or a non-stable product. Therefore, the parameters generate a particular geometry topologically, without allowing the matter to fossilise (Marcos 2010). This leads the products/objects to evolve and live in a trial mode, always ready to be altered according to the changing parameters (Carpo 2017). Similarly, living materials produce never-finished artefacts as a result of their metabolic activities, leading to morphological changes. They continually adapt to changing environmental conditions. Therefore, living matter can be perceived as an objectile. Subsequently, the following section delves into the concept of biological form generation and presents how this process can be likened to an algorithmic mechanism.

3. Biological Form Generation

In biology, the development of form is a complex process involving a combination of **genetic information** encoded in DNA, **mechanical forces**, and **physical and chemical environmental factors** (Dade-Robertson 2021). The genetic information encompasses details regarding cell shape, orientation, interactions with other cells, sensitivity to edges and forces, as well as inter-cellular communication (Dade-Robertson 2021). Cells use this information to organise their environment, which refers to the space around and between cells (Dade-Robertson 2021). Cellular systems can be engineered through synthetic biology techniques, providing the means to develop patterning, differentiation, and morphogenesis (Davies 2017). These techniques can be referred to as "*roads not taken by natural evolution*", offering insight into studies in evolutionary development (Davies 2017).

Mechanical forces are another influential factor in the generation of biological form. Biologist D'Arcy Thompson introduces an analytical perspective, showcasing the logic behind the transformation of biological forms over time. Thompson uses mathematical models to explain biological growth and form generation (Jarron 2014). He proposes that the form of a biological object can be explained by a "**diagram of forces**," whereby physical forces such as surface tension or gravitational forces play a role in shaping biological species or transitioning them from one into another (Thompson 1917). In the "theory of transformations", Thompson uses mathematical logic to point to transformations between biological species. Figure 3 demonstrates how scaling and shearing techniques can generate various fish shapes within the same Cartesian coordinate framework (Jarron 2014). Thompson's analysis illustrated the relationships between subspecies forms, which could transition into one another as in topological transformations.

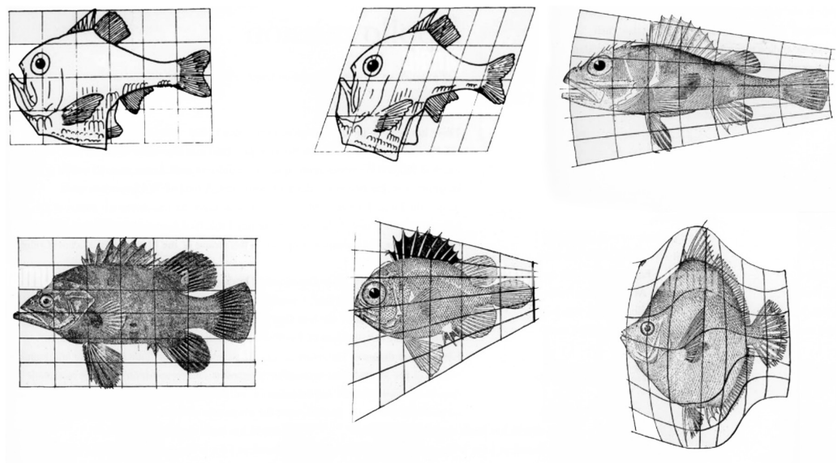


Figure 3

A drawing by Thompson that illustrates the transformations that are used to relate different fish to each other. Image from: https://www.alamy.com/stock-photo-darcy-wentworth-thompson-1860-1948-scottish-mathematical-biologist-31517404.html

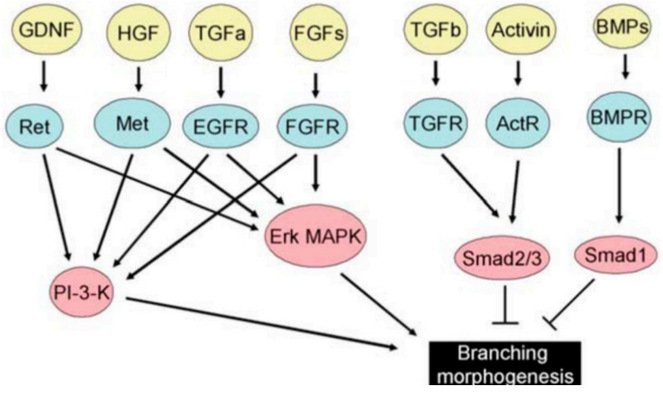


Figure 4

Pathway diagrams use arrows and boxes, where arrows indicate interactions between different components, such as genes and signalling molecules, and boxes represent stages or processes in development (Davies, 2023).

Thompson's "theory of transformations" is significant, as it pioneered the explanation of biological forms using mathematical models. His work formed the foundation for geometric morphometrics, which quantifies biological shape, shape variation, and covariation of shape with other biotic or abiotic variables or factors (Webster and Sheets, n.d.).

Environmental variables, alongside genetic information and physical forces, also have an influence on developmental pathways, which subsequently affect biological form indirectly. Environmental parameters include physical factors such as temperature, and humidity levels, as well as chemical factors such as nutrients. Scientist Jamie Davies describes **developmental pathways** as roadmaps that guide the formation and growth of an organism from its initial stages to its final form. These pathways often involve interactions between genes, promoters, and signalling molecules that regulate various aspects of development, as in Figure 4 (Davies, 2023). However, the process of morphogenesis is still not fully understood, therefore it is depicted as a black box in the pathway diagram (Davies, 2023).

In summary, the generation of biological form is a complex process influenced by both internal and external factors, including (i) genetic information, (ii) mechanical forces, and (iii) the surrounding physical and chemical environment. While genetic information and physical and chemical environmental factors induce indirect changes in biological form, mechanical forces or direct physical constraints can lead to direct effects (Figure 5). However, it is important to note that the biological object, in turn, has the capability to influence mechanical forces and environmental factors through counteractive forces or alterations in environmental conditions via the metabolic activities inherent to its growth. therefore, it is depicted as a dashed line in Figure 5.

4. Biological Parametricism

There is an analogy between parametric and biological processes in that in many examples of biological growth, especially in plants, the form of the organism is often, in part, derived from interaction with in vitro[3] factors, including access to sunlight and nutrients, physical constraints and barriers, and interaction with other organisms. To some extent, we already intervene in these biological processes in agriculture or gardening, by providing environmental regulation and irrigation such as a tomato, cultivated in a highly controlled, nutrient-rich environment with the use of robot arms, drones, and bespoke irrigation systems (Mazur 2016).

As Dade-Robertson mentions, refined crafts, such as the growing of Bonsai trees, are also examples of intervening in biological growth with specific forms in mind. However, while the cultivation of vegetables may lead to the production of high yields or larger individual crops, we do not intervene in the morphology of the tomato. The Bonsai tree, on the other hand, is produced through direct and cohesive control through the wiring of branches and roots and the severe limitation of nutrients to maintain the tree's dwarf form (Ozkan et al. 2022). From a design perspective, this cultivation approach is more akin to direct parametric control, operating on the form itself rather than indirectly influencing growth factors.

[3] In vitro methods explore how the effect of physical and chemical environment influences the biological form generation (Dade-Robertson 2021).

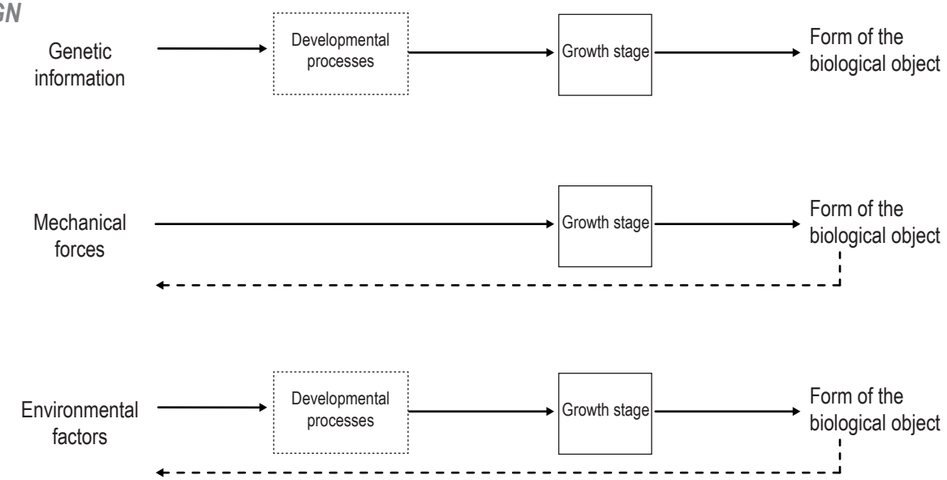


Figure 5

Diagram illustrating the distinct sequential process of interrelated factors converging to generate the biological form. A dashed line signifies that the biological object can impact mechanical forces and environmental factors through counteractive forces or alterations in environmental conditions.

This manipulation of form, achieved through direct constraint, resonates with the principles of D'Arcy Thompson's mathematical model for morphogenesis, which relates physical forces to the transformation of biological species.

On the other hand, biological forms can be indirectly influenced by environmental parameters that lead to different developmental pathways in growth, as described by Jamie Davies (Davies, 2023). The works of designers (Zhou et al. 2021, Zolotovsky 2017, Mitterberger et al. 2022, Groutars et al. 2022, Arnardottir et al. 2020) using plant roots, bacteria and silkworms show approaches to influence the environmental conditions of organisms to achieve desired material forms or functions (Table 2, Figure 6).

Table 2

A summary of the relationship between the material behaviour and its growth conditions.

Project	Organism	Parameters	Changes/ Outputs
Interwoven (Zhou et al. 2021) (Figure 6.1)	Plant roots	The growth media, direction of gravity and porosity, shapes, angles, and dimensions of their mould	The growth direction of roots
Guided growth of bacterial cellulose biofilms (Zolotovsky 2017) (Figure 6.1)	Cellulose-producing bacteria	Nutrients, substances, and airflow	Growth rate and formation
Degrees of Life (Mitterberger, Derme, and Imhof 2022) (Figure 6.2)	E. Coli bacteria	Deposition of nutrition	The growth rate and colour
Flavorium (Groutars et al. 2022) (Figure 6.2)	Flavobacteria	Temperature, humidity, and nutrients	Form, texture, and iridescent colour
Bacterial Sculpting (Arnardottir et al. 2020) (Figure 6.2)	Urease-producing bacteria	Cast sizes, inlet positions for nutrients and reactants	Form of the cemented sand

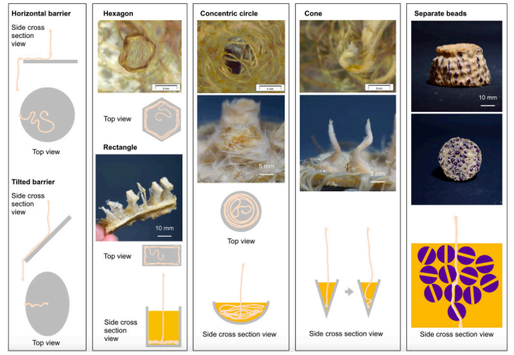
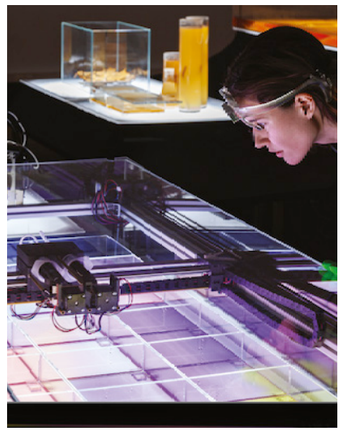
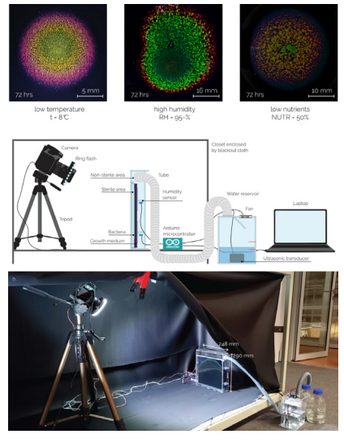


Figure 6.1

Interwoven (Zhou et al. 2021) and Guided growth of bacterial cellulose (BC) biofilms (Zolotovsky 2017).





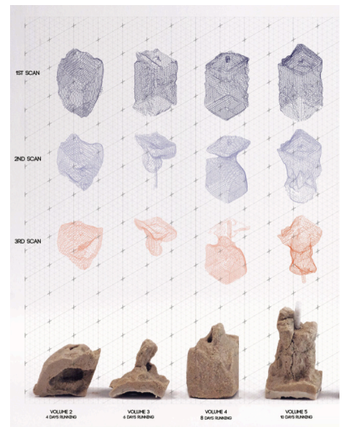


Figure 6.2

Degrees of Life (left) (Mitterberger, Derme, and Imhof 2022), Flavorium (centre) (Groutars et al. 2022), and Bacterial Sculpting (right) (Arnardottir, Dade-Robertson, and Mitrani 2020).

Zhou et al. used plant roots to test digital biofabrication strategies for product design purposes (Zhou et al. 2021). They fabricated self-supported 3D structures by altering the growth media, direction of gravity, and porosity of their mould. These variables allowed them to manipulate plant roots, since the nutritional richness and the force of gravity have an impact on the root growth (Zhou et al. 2021). Similar to the bio-digital fabrication of plant roots, Zolotovsky's 'guided growth' of cellulose-producing bacteria demonstrated ways to program bacterial cellulose as a living material that responds to the environment. Her system computationally adjusts the flow of nutrients and adds substance to help the designer interact with the biological process of growth and material formation (Zolotovsky 2017). Milterberger et al. explored the interaction between humans and E. coli bacteria using a sensor system (an eye-tracking system) that activates different interaction routines according to defined rulesets (processed by machine learning) (Mitterberger, Derme, and Imhof 2022). As a result, human gaze interaction controlled the deposition of nutrition at specific locations, which then affected the growth rate and the colour of the microbial mat. In the Flavorium project, the form, texture, and iridescent colour of Flavobacteria were modified through environmental stimuli, including low temperature, high humidity, and nutrient deficiency (Groutars et al. 2022). The response of Flavobacteria allows the designers to tune growing conditions in a more controlled manner using digital systems. Although Arnardottir's fabrication process did not include digital tools in controlling the parameters, she used urease-producing bacteria to calcify sand, creating cemented columns of material (Arnardottir et al. 2020). By altering the cast sizes, and inlet positions for nutrients and reactants, Arnardottir demonstrated that parameters which affect biological growth can be influenced. The influence of the parameters could be predicted while creating cast materials; the final form of the cemented columns did not have to be dictated by the shape of the cast (which is a direct physical constraint). More complex forms emerged because of the interaction of these biological and environmental factors.

As seen in each of the examples above, biological organisms continuously interact with their environment and transform their state, morphology, behaviour, and performance through their metabolic cycles, life spans and growth rates (Thomsen 2022). Environmental changes act as input variables, prompting changes in the biological objects. Designers initially define these environmental factors for biological computation in a parametric manner, enabling them to fine-tune material properties and influence the outcome. In a way, these in vitro factors create boundaries or constraints for the organism, offering varying levels of control that can be implemented digitally or manually.

In summary, biological forms can be influenced directly, as in the topological design model, by mechanical forces such as Bonsai shaping. Alternatively, they can be formed through associative processes, such as environmental changes or genetic modifications, resembling the associative design model (Figure 7). The varying directness or indirectness of these input-output relations also share commonalities with the dynamic design model, as the growth of the biological objects occurs in stages over time, introducing a temporal dimension to the generation processes of biological forms.

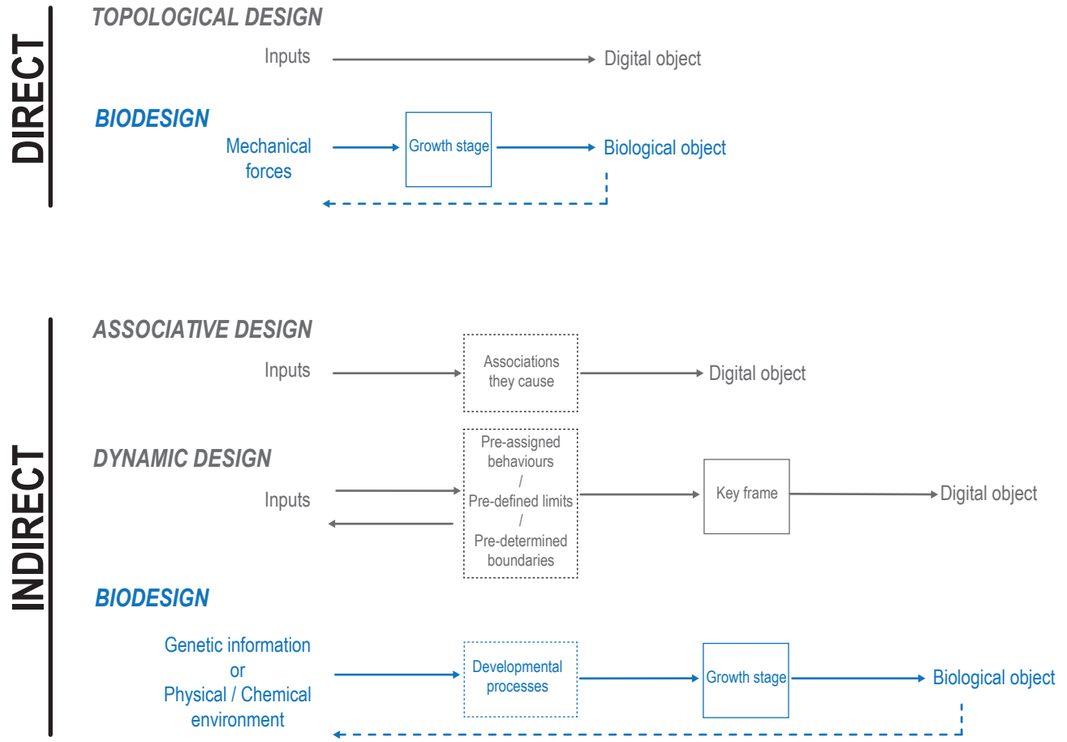


Figure 7

In digital formation models, the designer interacts with computational generative mechanisms that influence the form of the object accordingly. This same logic applies to biological form generation. The diagram compares digital and biological form-generation processes.

5. The Design Space

Although there may be commonalities in the way digital and biological forms respond to stimuli or inputs, there are also distinct characteristics that set them apart. While digital objects form within topological space, biological form conflates two types of space: one which is 3D space and the other which describes a phase space. Understanding where the form of an artefact is generated holds significant relevance in design, as it allows designers to move beyond the constraints of the conventional rigidity of Euclidean geometry and explore new possibilities for form-making (DeLanda 2005). Additionally, understanding the various types of design spaces equips designers with additional tools for deciphering complex systems and generating diverse forms (Cogdell 2019).

Phase space and topological space differ primarily in their levels of computation. **Phase space** is a comprehensive concept encompassing all potentials, capacities, and possible states or phases of a system (DeLanda 2005). It provides a more holistic view of the behaviour of living systems, enabling exploration of their evolution and changes over time. On the other hand, **topological space** provides a way to study the structural possibilities in mathematical models (DeLanda 2012). Within this topological space, where curvilinear forms exhibit instability and fluctuations, new potentialities, such as emerging forms, come to the forefront (Kolarevic 2001). Each emergent geometry in topological space is a variant of the original model. In this regard, the key lies in surface connectivity. For example, in the case of a blob's subdivision surfaces, the topology remains unchanged even if the designer adds or removes subdivisions. As seen in Figure 8, the blob can transform from a doughnut shape to a cup shape through the application of forces like stretching, folding, and twisting. In digital formation models, digital objects can take a wide array of shapes in Euclidean geometry, while preserving their essential topological similarity.

Topological space provides a single calculated result based on a set of mathematical models without accounting for all possible outcomes or implications formed by the designer and software. In contrast, phase space requires more complex calculations since it encompasses all possible states and interactions within a system. Biological objects can represent multiple states in phase space, reflecting their complex and dynamic nature. The environmental variables or mechanical forces that influence the biological form are mapped with relationships in multiple dimensions. The organism's response is shaped by multiple factors, including its internal processes, environmental conditions, and interactions with other systems. Every small thing can have a significant impact on the overall morphology, leading to a wide range of possible outcomes. An organism's responsiveness makes the outcomes highly varied and dynamic.

How a cell interacts with its environment to organise gene action appropriately and responsively is the subject of epigenetics (Cogdell 2019). The change in the genetic level of a cell through alteration of the environmental conditions occurs in the **epigenetic landscape**. Based on Weddington's idea, Dade-Robertson defines phase space as a multidimensional space, "the dimensions of which are based on some property of the object they represent"

Figure 8

A model demonstrating a topological transformation of a mug to a torus

Image from: https://tomrocksmaths.com/2022/07/12/teddy-rocks-maths-essay-competition-2022-overall-winner/.

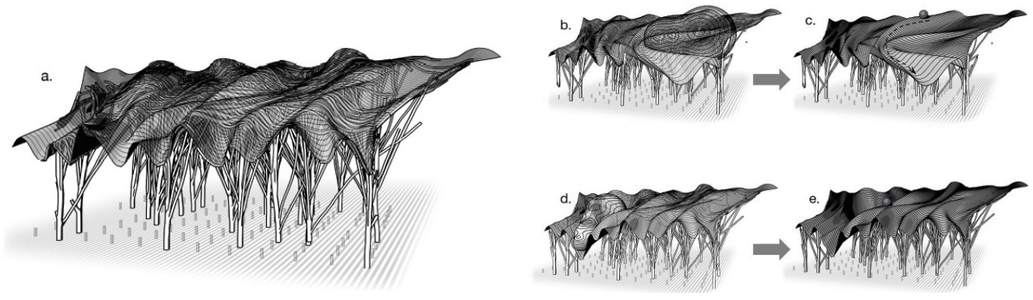


Figure 9

A revised version of developmental biologist Waddington's developmental landscape by Dade-Robertson showing the external influences on the 'creode' or necessary path of cell development.

(2021). The variables of the phase space are time and conditions such as temperature, humidity, or CO_2 level (Dade-Robertson 2021). In phase space, cells, as fundamental structural and functional units of organisms, undergo growth and transformation over time. This transformation leads to the concept of a “creode” representing the unique path a cell takes as a result of environmental changes over time (Dade-Robertson 2021). **Creodes** are shaped by interactions between various biochemical processes and environmental factors and are specific to each cell (Dade-Robertson 2021). Exploring creodes offers new insights for researchers and designers into the behaviour of living systems, and develops new methods for creating functional and adaptive designs.

In Figure 9, the gene expression of a cell is represented by columns, and the overall geometry or the paths that the cells follow are illustrated by the surface on top of the columns. Altering environmental conditions changes the paths, resulting in alterations in the epigenetic landscapes represented by the surfaces (Dade-Robertson 2021). In turn, it prompts corresponding changes in cell behaviour, as seen in panels b, c, d, and e, through variations in column heights.

Consequently, the concept of design space matters because it provides a framework for understanding and exploring the creation of forms across diverse domains, ranging from digital design to complex biological systems. By exploring various types of spaces, designers can transcend conventional constraints and unlock new possibilities. In this context, applying a parametric approach, akin to existing in topological space, within the realms of epigenetics and design creodes, can facilitate many advantages. This structured, parameter-based methodology serves as a tool for understanding complex epigenetic processes and using them to refine qualities while biofabricating. Instead of prescribing the final form or qualities of biological objects from the start, it provides the space for the living material to perform, and compromises its inherent qualities.

6. Conclusion

As living materials are new and complex systems in the architecture field, they necessitate a new approach to the interaction between organisms and the fabrication processes (Thomsen 2022). This chapter has explored the possibility of altering the developmental pathways of biological systems through the application of associative design principles. These principles involved an analysis of topological, associative, and dynamic modelling methods found in relevant literature. As these digital design processes are non-deterministic, they enable designers to actively engage within form-generative environments (Oxman 2006).

This chapter compared the processes involved in creating biological and digital artefacts, highlighting both their similarities and distinctions. The analysis concluded that digital formation models use highly defined direct or indirect associations, whereby each input value (such as forces applied or numerical inputs) is distinctly represented by a morphological output. However, within biological systems, the translation of them is more indirect or nonlinear, while a correlation still exists. This complexity arises from the nature of biological systems, which possess multiple hidden layers beyond the designer’s immediate perception. In digital formation models, the algorithms can be unpacked or changed by the designer, providing a degree of transparency. In contrast, biological generation processes function more as enigmatic “black boxes” within the organism’s metabolism, challenging straightforward manipulation and understanding.

After classifying various types of digital formation approaches, the next chapter focuses on whether it is possible to use parameters and build indirect associative relationships with the developmental pathways of living material in the design and fabrication processes. The core objective of Chapter 5 is to explore the possibility of utilising a digital formation approach for designing and producing living materials. Consequently, a series of design experiments was conducted to address this question. These experiments led to an understanding of whether a biofabrication process can be characterised as generative or associative.

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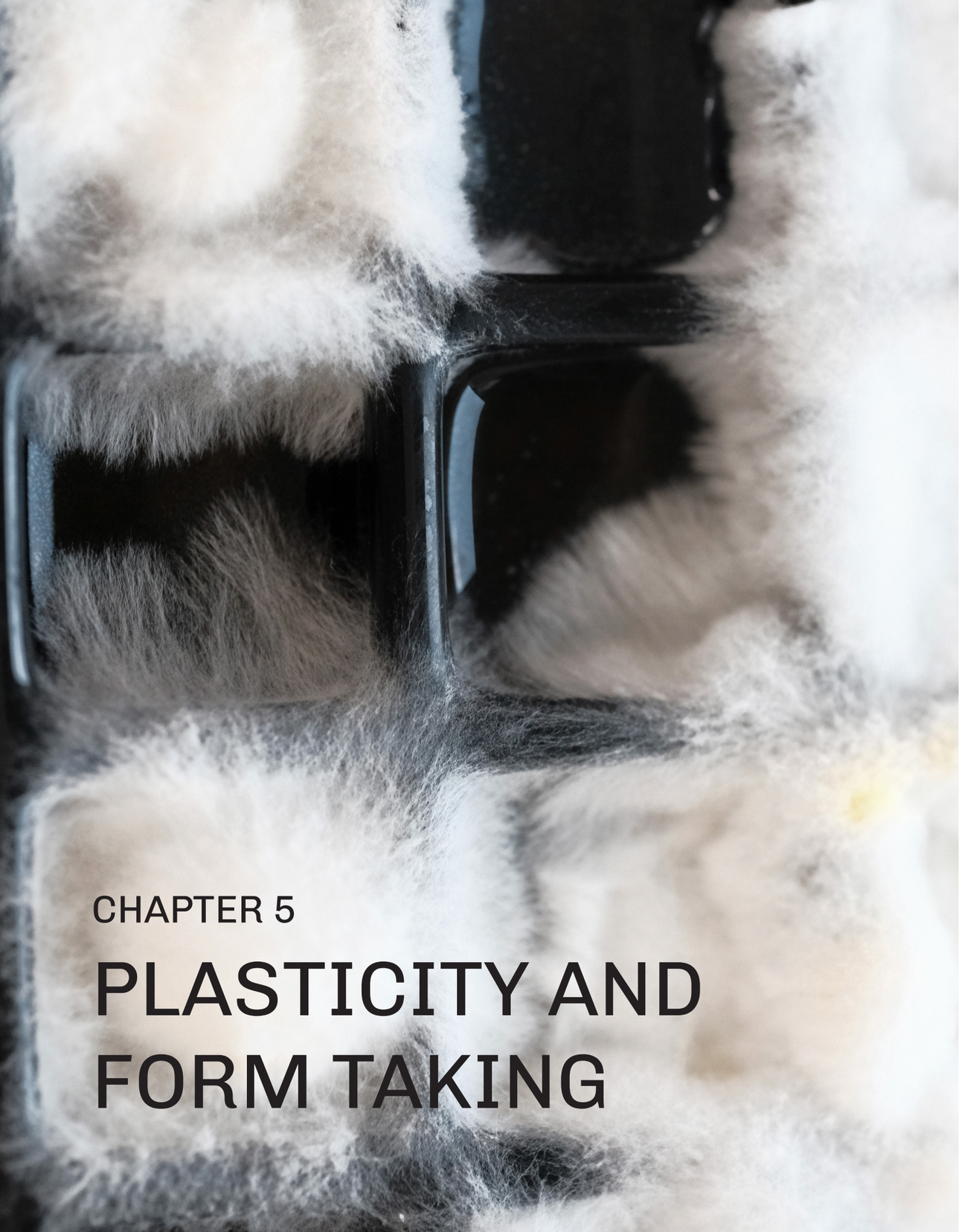


CHAPTER 5

PLASTICITY AND FORM TAKING

1. Introduction

One of the primary goals of this thesis is to establish a biofabrication method that (1) is applicable to making-through-growing, (2) facilitates material interaction, and (3) allows designers to work with the tendencies and capacities of living materials. To realise this vision, the previous chapter introduced the concept of a parametric approach for fabricating living materials. A parametric approach enables the application of methods from digital formation models to biodesign, and helps designers address the complex nature of biological systems. This, in turn, allows designers to tune material qualities through input variables and create bespoke forms or functions (akin to digital formation models). Consequently, the main objective of this chapter is to explore how the fungus can be used as a biomaterial probe to test the concept of a parametric approach as a biofabrication method developed in terms of New Materialism.

To define biological processes as parametric, a comprehensive study of fungi was conducted, with a particular focus on their growth and morphological development in both their root network (hyphae/mycelium) and fruiting body (mushroom) forms. The initial objective was to understand the characteristics of fungal growth, and the factors influencing it, as well as to identify constraints (if any) on the various stages of its growth. These questions also led to investigating the concept of plasticity, in both philosophical and biological contexts, aiming to answer how fungi form. Developmental plasticity is a characteristic of living systems that demonstrates the tendency and capacity of a material to undergo change or adaptation. Subsequently, after defining the factors influencing fungal development from the literature, a series of design experiments was initiated (as in Sections 5.1 and 5.2), starting with mycelium. These early design experiments aimed to influence the organism's morphology through its adaptability and developmental process. These experiments were conducted utilising craft-like processes. The primary parameter examined was nutrient levels, a chemical environmental factor affecting mycelium properties and behavioural patterns. The underlying goal was to examine the relationship between a specific environmental factor and the morphological changes as mycelium adapts to varying chemical environments. The concept involved locally altering the environment of aerial mycelium that extends in the air (Kuribayashi et al. 2022a), by the deposition of diverse nutrients. Given the understanding that the organism morphs on a micro-scale, resulting in the creation of various geometries overall, as shown in Figure 1 in response to various nutrients, the objective was to leverage this developmental plasticity as a biofabrication method.

Chapter 5 *is reworked from the conference proceeding D. Ozkan, M. Dade-Robertson, B. Christgen, Demonstrating a Material Making Process Through the Cultivation of Mycelium Growth, in 4th International Conference for Biodigital Architecture & Genetics, 2020.*

This process also had the potential to influence material properties, including density, strength, and permeability. In a way, it was conceptualised as a 3D irrigation system or printer, sharing similarities with these machines in that they both follow computer commands to run bio-ink or solution according to given instructions. There was a key difference, as the 3D printers work with materials as a homogenous paste for extrusion, whereas this approach aimed to leverage the inherent qualities of the biomaterial to facilitate autonomous growth. That means that the adjusted variables would guide (push and pull) the aerial mycelium formation towards a desired outcome without direct physical intervention.

2. Plasticity

The decision to select fungi as a material probe was significantly influenced by plasticity. **Plasticity** has various meanings in biology, physics, and materials science. In philosophy, it means “the capacity of the living being to receive form and to give form, and also the capacity to explode form” (Zukauskaite 2023). Within a biological context, plasticity is “the capacity of organisms or cells to alter their phenotype in response to changes in their environment” (Skipper, Weiss, and Gray 2010). This adaptation is crucial, as it enables observable alterations to occur within the same genes, without mutations or long-term genetic shifts. **Developmental plasticity** leads to variations in phenotypes based on the same genotype, depending on the environmental conditions throughout the organism’s developmental process (Lafuente and Beldade 2019). The generation of diverse phenotypes influenced by environmental factors (Skipper, Weiss, and Gray 2010), involves **epigenetic processes**, enabling organisms to adapt. This relationship between phenotype and environment can be measured in terms of the degree of variation among organisms with the same genome (Dade-Robertson 2021). For instance, plant leaves alter their shape in response to factors such as light, temperature, and water, as a result of their developmental plasticity (Figure 2) (Nakayama, Sinha, and Kimura 2017).

Developmental plasticity can take place at various stages during embryonic development, operating at different levels, ranging from individual cells to the organism (Skipper, Weiss, and Gray 2010). The conventional scope of developmental plasticity typically encompasses the entire organism, involving psychological and behavioural plasticity. However, this thesis specifically delves into a distinct subset—morphology. It focuses on morphological diversifications, exploring the intricate aspects of form and structure caused by changes in physical and chemical environmental conditions, rather than genetic alterations. These phenotypic changes happening in the form are also known as **morphological plasticity** and refer to species-specific adjustments induced by environmental changes (Sultan 2003).

As illustrated in Figure 3, achieving a particular morphology in biology involves a developmental process. Individual cells undergo this process and some transition into multicellular entities through the activation of a single genotype’s capacity and its associated developmental pathways (Moczek et al. 2011). The developmental process continues until a point where significant changes in the capacity for further development become limited. This capacity to make specific functional adaptations in response to the environment, including distinct developmental, physiological, and reproductive alterations, is called **adaptive plasticity** (Sultan 2003).

Dade-Robertson explains that the level of development plasticity within organisms can fluctuate throughout their lifetime, depending on factors such as their survival rate, mobility, adaptability speed, and the extent of their adjustments. While there is no precise measure or scientific distinction regarding the level of plasticity, it can be observed that some organisms (or even the same organisms in specific states), exhibit a greater degree of adaptability compared to others. They all adapt to their environment in diverse ways to ensure survival,

Figure 1

Assumed aerial mycelium growth in various forms with nutrient deposition.

Figure 2

Top view of shoots grown under each condition for a month. Left, at 20°C; right, at 30°C. (B) Mature leaf morphology of the seventh leaf. Left, at 20°C; right, at 30°C. Bars = 3 cm in (A) and 2 cm in (B) (Nakayama, Sinha, and Kimura 2017).

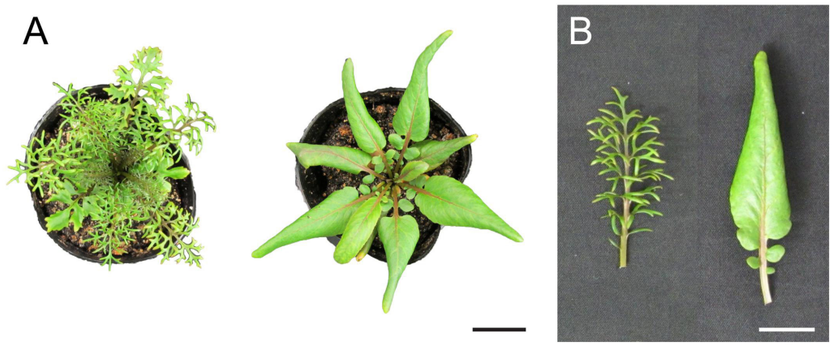
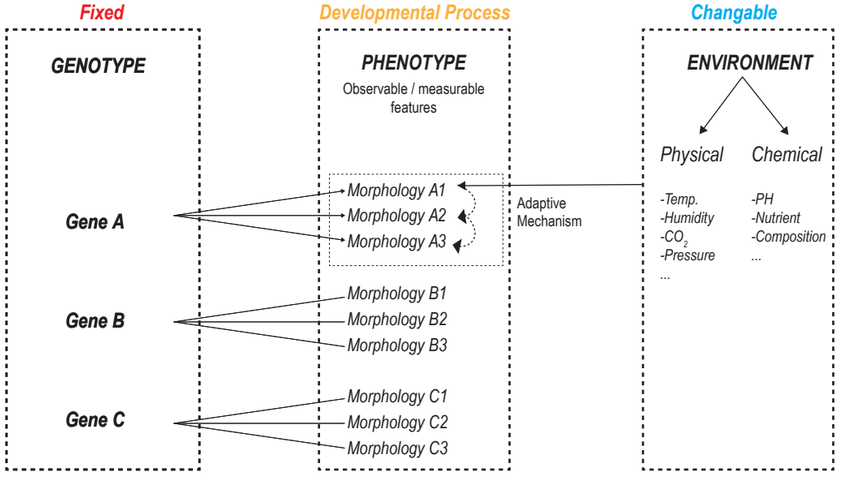


Figure 3

A diagram showing the relation between genes, morphology, and environmental factors.



through abilities like movement or morphological adjustments. In this thesis, the terms “**low plasticity**”, or “**high plasticity**” are used, reflecting the degree of developmental sensitivity and phenotypic changes observed in the organism, aligned with its environmental conditions. In this context, as Dade-Robertson explains, plants demonstrate a higher degree of plasticity compared to humans (Dade-Robertson, supervision meeting, November 1, 2023). Plants in high or low environmental humidity can alter both their leaf and root-absorptive surface areas in a short time. This implies that phenotypic response can occur in different parts of the same organism (Sultan 2003). In contrast, organisms such as humans may exhibit a relatively limited degree of plasticity, as the morphological changes are less conspicuous than in plants.

This thesis explores the developmental plasticity of fungi as a biofabrication method within a New Materialist approach. Although the primary focus is on *Pleurotus ostreatus* (oyster mushrooms), the principles underlying these changes are likely applicable to other fungal species or living materials, as well. Through the modification of specific environmental factors, the organism’s morphology can be altered (up to a certain extent), providing a method for designing with living materials. This allows for making-through-growing and influencing the material’s morphology without any direct physical interaction, relying only on environmental manipulation. Furthermore, these environmental variables can be considered as precisely controllable inputs, influencing the organism’s growth as outputs. This relationship can be systematically established in a parametric manner. By adjusting the environmental inputs, the output morphology can be fine-tuned within the plasticity limits of the organism. As a result, there is a need to understand the characteristics of fungi and the factors influencing their growth and development. To identify any constraints, the growth stages of fungi, initially in mycelium form, were explored through literature.

3. Fungal Mycelium

3.1. A General Overview of Fungi

There are approximately 1.5 million fungal species in the world (Moore 2010). They are ubiquitous, even being found in the air as spores. They take their origins as a distinctive group of unicellular eukaryotes, although they exhibit different properties from each other (Chang and Miles 2004). Plants and animals also have eukaryotic cells, but fungi differ from them with their cell wall composition. Fungi cell walls are unlike animals and plants (Chang and Miles 2004). Their main role in nature is to decompose and cycle elements such as carbon, nitrogen, and oxygen (Chang and Miles 2004).

Fungi mainly consist of two parts, the fruiting body (the mushroom) and the mycelium (the roots) (Figure 4). **Mycelium,** the root system of fungi, is composed of hyphae, which are tube-like fibres with a diameter of approximately 10 µm (Islam et al. 2018b). Mycelium is part of fungi that is of general interest as a biomaterial, as it can be grown rapidly on various forms of waste, creating a bulk building material (Islam et al. 2018b). However, this does not mean every mushroom-forming fungus, such as portobello, button, and porcini mushrooms, can be used to produce useful biomaterials. The fungus types most often used in biomate-

Figure 4

Parts of fungus.

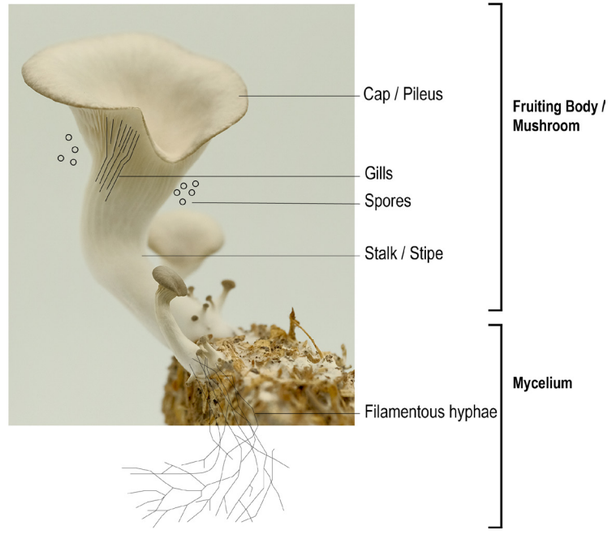
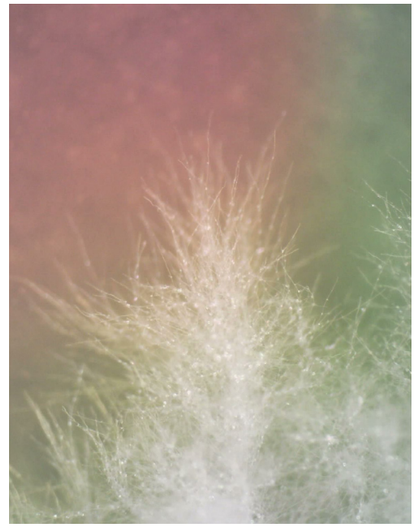


Figure 5

Microscopic images of mycelium (Dino-Lite digital microscope at 70X magnification).



rial production are wood decay fungi which can degrade cellulose, hemicellulose, and lignin, biopolymers found in the cell walls of plants (Appels et al. 2019). They form filamentous mycelium, and they belong to the *Basidiomycota* group of the fungal kingdom. The enzymes of these fungi work to break down these biopolymers into substrates (Rocco and Kalisz 2012). A filamentous fungus can therefore live in geometrically, mechanically, and materially heterogeneous environments that are rich in degradable cell walls, such as plant tissue, decaying wood, leaf litter, and soil (Held et al. 2019). While forming mycelium, as well as an organic carbon source such as decayed cell walls, a nitrogen source (e.g., peptone, yeast, or beef extract) is also required (Watkinson, Boddy, and Money 2016).

Mycelium can be produced in different forms, such as **pure-mycelium** (mycelium leather, aerial-mycelium) and **mycelium-composites**. Pure mycelium materials and mycelium leather are solely made from mycelial biomass, showcasing various properties (Vandelook et al. 2021). (Vegetative) aerial hyphae, extending into the air, contribute to the formation of aerial mycelium from pure mycelium biomass (Kuribayashi et al. 2022). Mycelium composites consist of networks of filamentous hyphae, utilising biological growth to transform low-cost organic wastes into economically viable and environmentally friendly materials (Jones et al. 2017).

While mycelium is relatively simple, the f**ruiting bodies**, which are composed of cells with the same genetic code as the roots, are morphologically complex with a high degree of cell differentiation. The fruiting bodies are formed by a **stipe** (stalk) supporting a **pileus** (cap) where the **spore** production occurs (Watkinson, Boddy, and Money 2016) (Figure 4). The spore production, which helps to transmit fungi through space, takes place under the mushroom caps in the **gills** (Sakamoto 2018).

3.2. Fungal Primordium

A **hypha** extends from its tip, and branches out as it grows (Figure 5, right). When it meets with another hypha they come together and form a complex multi-fibrous network structure (Figure 5, left) (Islam et al. 2018b). Hypha filaments can keep growing up to a certain point by seeking new carbon sources, even without any available source of nutrition (Held et al. 2019). Their cell wall is composed of chitin nanofibers, beta-glucans, and an outer layer of proteins (Haneef et al. 2017). The hypha wall is the main constituent of their structure and has an impact on fungi morphogenesis. It helps to conserve and supply mechanical stability to hyphae filaments (Haneef et al. 2017).

Hyphae filaments have various structural qualities according to the species of fungus. They can have different characteristics in terms of texture, density, and colour (off-white, white, or pale pink) (Masoumi et al. 2015). For *Basidiomycota*, there are three main hyphal types; generative, binding, and skeletal (Jones et al. 2017). These types help in determining the mycelium strength and, therefore, the material's mechanical performance (Jones et al. 2017). Hypha species can possess one (monomitic), two (dimitic) or three (trimitic) hypha types. A monomitic species can provide limited strength, while dimitic and trimitic species are stronger. For example, a trimitic species such as *Trametes versicolor* (Turkey tail) has a higher compressive strength than a monomitic species such as *Pleurotus ostreatus* on the same growth medium (Jones et al. 2017).

4. Influencing Mycelial Growth

Before conducting design experiments aimed at going beyond the limitations of moulding techniques and fabricating mycelium in a parametric manner, different methods were explored to fabricate mycelium materials and influence its growth without forcible constraints, that align with a "making-through-growing" approach. Each piece of research presented in this section worked with fungi and focused on understanding their behaviour for diverse applications. By investigating the methods and outcomes of these studies, we, as designers, aim to gain knowledge that serves as a foundation and inspiration for working with fungi. Additionally, these examples help to identify the parameters that can be used to influence mycelium growth in design experiments. Although each method serves a different purpose, collectively, they provide design inspiration by demonstrating ways to guide the growth of fungi with minimal physical intervention.

These methods modify specific environmental factors to influence the mycelium's morphology, density, growth direction and material properties accordingly (to a certain extent). Environmental factors can be considered as precisely controllable inputs that influence the organism's growth as outputs. This relationship can be systematically established in a parametric manner. By adjusting the environmental inputs, the output growth characteristics can be fine-tuned within the organism's plasticity limits. Therefore, understanding the characteristics of mycelium and the factors influencing its growth and development is essential. Techniques to influence mycelium growth in both pure mycelium and mycelium composites can help with (i) adjusting mycelium density, (ii) altering growth direction, (iii) modifying hyphal morphology, and (iv) forming hyphal knots. Table 1 summarises research on the effects of electric spiking, photomasking, chitosan treatment, and fed-batch fermentation on mycelium formation, highlighting the parameters that can be adjusted to influence mycelium growth in experimental design. Although each technique serves a unique purpose, collectively they offer design insights by demonstrating methods to guide fungal growth through their agency.

Adjustments in mycelium growth parameters lead to variations in its material properties, allowing it to adapt to diverse functional outcomes based on fabrication conditions. This adaptability makes mycelium a self-fabricating system, where changes in growth parameters trigger variations in its material characteristics. The potential for versatility is due to the plasticity of mycelium, which allows it to be a self-regulating material and to respond dynamically to environmental changes during growth, expanding its field of possibilities and enabling a diverse array of properties.

Among the methods listed above, two will be the focus, as these methods are considered particularly applicable for this PhD: the use of nutrients and the inclusion of other species. A more detailed description of these methods is provided below.

Table 1

A list of methods influencing mycelium growth.

To	Methods	Description
Tune mycelium density	Temperature change	For certain fungal species hyphal density and aggregation count are temperature sensitive, with branching patterns varying across different temperatures in both solid-state and liquid fermentation (Reynaga-Peña and Bartnicki-Garcia 1997). When the temperature increases, hyphae at the edges of fungal colonies in petri dishes begin forming new branches at their tips (Reynaga-Peña and Bartnicki-Garcia 1997).
	Electrical current	Fungi modulate electrical activity as a response to environmental stimuli. Mycelium, for instance, reacts to pressure or humidity changes by generating electrical spikes, allowing it to process information within its network (Adamatzky, Gandia, and Chiolerio 2021). This suggests that inducing an electrical current could alter mycelium behaviour by influencing its network communication. In addition to the electrical spiking initiated by hyphae, electric current or the application of electrical stimulation can significantly impact mycelium growth (Majib et al. 2024). Studies indicate that low levels of DC and AC currents applied through wire electrodes in agar can increase mycelium growth rates in petri dishes (Jamil et al. 2018).
	Heterogeneously distributing nutrients	The level of nutrients affects mycelium density (Ritz and Crawford 1997). Their work shows that using a tessellated agar tile system with a grid structure and heterogeneous nutrient distribution results in varying levels of mycelial density and extension.
	Repeated batch fermentation	Repeated-batch fermentation (RBF) periodically replaces part or all of the liquid medium with fresh medium, without disturbing the ongoing culture, leading to higher mycelium production than the batch method (Wan-Mohtar, Ab Kadir, and Saari 2016). The composition of fungal cultures changes during fermentation, influencing the production of polysaccharides, which correlate with mycelium density. Higher mycelium density boosts polysaccharide and ganoderic acid production, altering fungal cell size and shape (Wagner et al. 2004). Thus, manipulating the medium composition in RBF helps regulate both mycelium density and morphology.
Alter growth direction	Chemical signalling	The growth direction of hyphae can be influenced by attracting or repelling them, guiding their movement and the location of clamp connections in filamentous fungi through hyphal self-signalling and self-fusion. This self-signalling mechanism between hyphal tips can be influenced by chemical compounds (Read et al. 2009).
	Through other species	One fungal strain can dominate and overtake others (Hiscox, O'Leary, and Boddy 2018). The antagonism observed between different mycelial species could be used as a method for controlling growth in a designer-specified manner. Theoretically, modulating the amount of the dominant strain could regulate the growth of the recessive species as desired (Crowther, Boddy, and Maynard 2018).
Modify hyphal morphology	Chitosan or chitin treatment	Hyphae cell walls contain chitin, and changing the protein content in these walls affects mycelium morphology (Oliveira Junior 2016). Modifying chitosan, derived from chitin, through various methods, can further impact its properties and effects. For example, removing acetyl groups strengthens chitosan, enhancing its effectiveness (Oliveira Junior 2016). Adding calcium chloride is another approach that can modify chitosan's interaction with fungal cells (Vandelook et al. 2021).
	Feedstock type	The properties of mycelium in composites depend on variables like nutrient substrates and substrate size, which influence hyphal characteristics. For example, enriched feedstock can alter hyphal thickness or structure (Haneef et al. 2017). Mycelium becomes stiffer when consuming pure cellulose or other hard-to-digest substrates due to increased chitin synthesis, whereas it becomes more elastic with dextrose-rich substrates (Haneef et al. 2017).

I. Through nutrients

There are numerous studies on the quality of mycelium composite materials, including research by Appels (Appels et al. 2019), Jones (Jones et al. 2017), Elsacker (Elsacker et al. 2019), Attias (Attias et al. 2017), Islam (Islam et al. 2018a) and Haneef (Haneef et al. 2017). Their work is centred on altering the properties of mycelium composite materials, often by modifying factors like nutrition substrates, substrate size, and time as variables. These variables then influence mycelium behaviour and hyphae properties, consequently impacting the composite material's quality. For example, Haneef et al. enriched one nutrient source and compared the results with a sample without enrichment (Haneef et al. 2017). As seen in Figure 6, the mycelium thickness remained consistent from the 5th to the 20th day in both samples, yet there was a notable difference in structure. It showed that the thickness or structure of hyphae remained nearly the same over time, and the feeding substrate affected the filament thicknesses in every stage of growth (Haneef et al. 2017).

It was also discovered that mycelium becomes stiffer when it consumes pure cellulose or any substrate that is difficult to digest due to the increase in chitin synthesis. On the other hand, it becomes more elastic when dextrose is consumed as part of the substrate. In short, as a common feature of all hyphae, the thickness of filaments is **time-independent** and **substrate-dependent** (Haneef et al. 2017). This experiment shows that mycelium can form various characteristics, such as stronger and weaker natural polymeric composites, depending on the arrangement of the nutrient substrates.

Another method to understand how to affect the growth and density of pure mycelium was developed by Karl Ritz and John Crawford (Ritz and Crawford 1997). They set up an experiment in a tessellated agar tile system by distributing nutrients heterogeneously and growing mycelium on the nutrients. They aimed to predict the behaviour of mycelium after analysing and decoding its development pattern. Understanding the formation of mycelium allowed them to produce mathematical models of its behaviour. Those models were applied in dynamic simulations and allowed their users to develop foresight by examining the potential

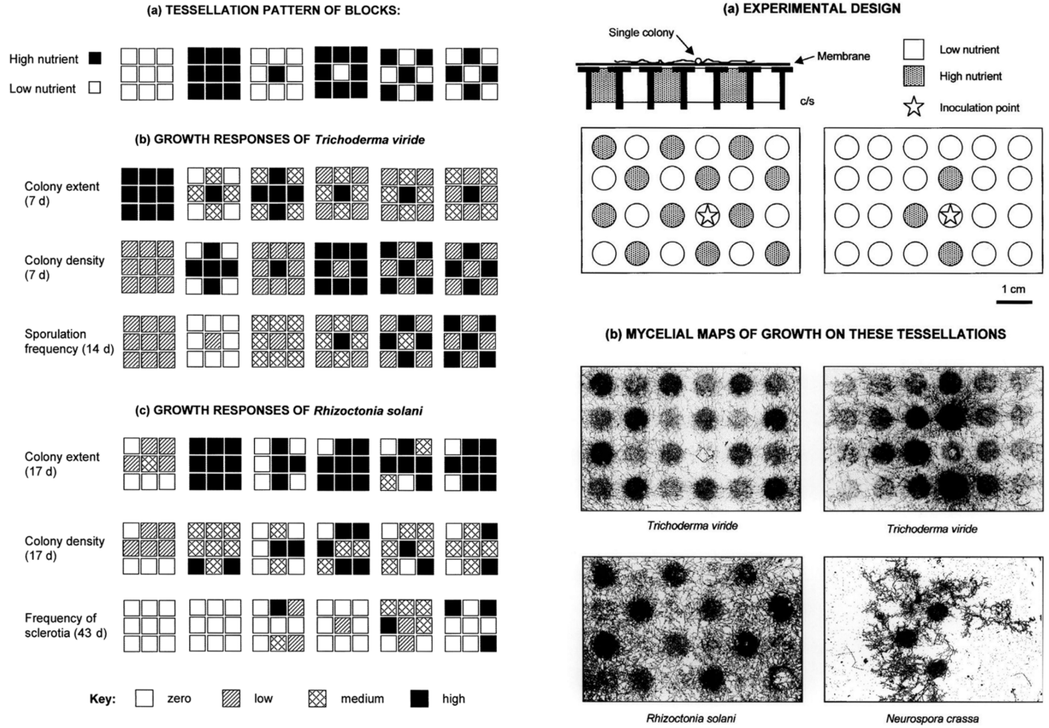


Figure 6

Tessellated agar spot system for producing large-scale mycelial maps of fungi growing in nutritionally heterogeneous microcosms (Ritz and Crawford 1997).

consequences and interactions of the organism under changing circumstances (Ritz and Crawford 1997). This work from Ritz and Crawford shows the principle of nutrient levels affecting the density of mycelium, as seen in the lower left corner of Figure 6. The use of a grid structure allowed them to set up experiments in a repeatable, systematic manner and provided results giving scope for quantitative analysis.

The examples show that altering nutrients can be used as a variable to change the mycelium qualities such as density, hardness, elasticity, and durability. Therefore, nutrients are set as a parameter in the experiments that could potentially affect the mycelium morphology.

II. Through other species

Another approach that influenced mycelium growth was applied by Thomas Crowther, Body and Maynard to understand the complexity of fungal systems (Crowther, Boddy, and Maynard 2018). They used artificial media to grow different mycelium species and studied the patterns, which they used to form a model to understand real-world fungal ecology, as seen in Figure 7. This helped them to identify, classify, and see the potential of various strains.

The initial research on the interaction of different fungal species formed the groundwork for the ‘Fungus Wars’ study by Hiscox, O’Leary, and Boddy (Hiscox, O’Leary, and Boddy 2018). The Fungus Wars study focused on understanding the mechanisms underlying wood decay *Basidiomycete* community dynamics and their decomposition processes for modelling ecosystem function, as seen in Figure 8 (Hiscox, O’Leary, and Boddy 2018). They used 2 to 27 wood cubes with pre-colonised mycelium to study the hyphal interactions of different species. It was observed that there was an antagonistic interaction between fungal communities when they were decomposing woody resources.

These studies illustrated that one fungal strain tends to be dominant and overtakes the others. The antagonism demonstrated between different mycelium species could be taken forward as a way of limiting the growth in a manner desired by the designer. The dominant strain could be used to create the boundaries for the recessive fungal species. Theoretically, modulating the amount of the dominant strain could affect the growth of the recessive species as desired. Although conceptually this method appears to be a good solution for controlling the growth of the dominant species, it would present its challenge.

In summary, using a tessellated agar system and a dominant second strain are both methods which have the potential to be employed to change the density and to restrict mycelium growth in a controlled manner. Both methods were considered for use in this thesis; however, the Fungal Wars concept was not utilised. This approach, although interesting, was not taken forward, as it requires more than one species of mycelium to be grown. Not only would this cause the challenge and cost of growing more than one mycelium species, but it would also make the growth dependent on another unpredictable living organism rather than more controllable environmental variables. As a result, the method from Ritz and Crawford was chosen to be taken forward into the creative practice.

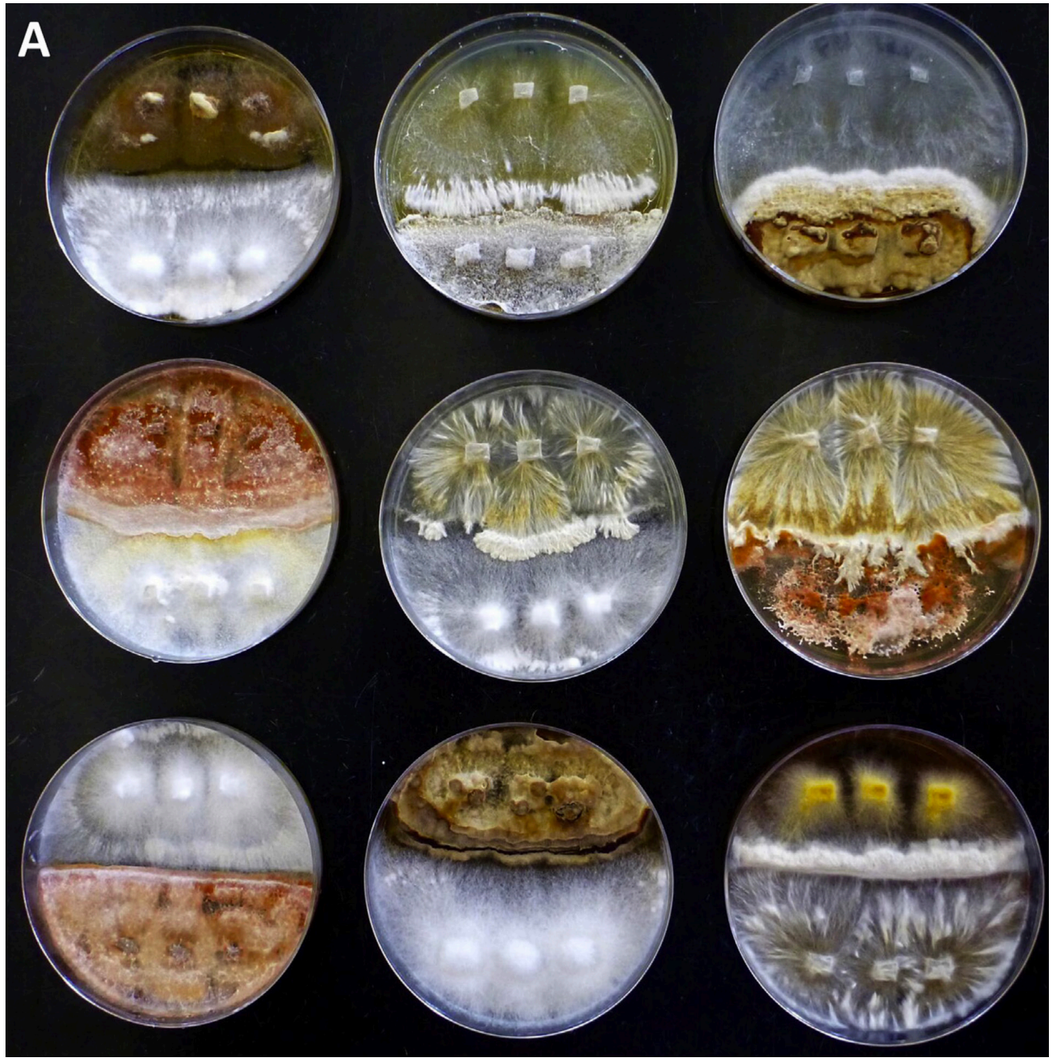


Figure 7

A range of fungi used in pairwise competitive interactions (Crowther, Boddy, and Maynard 2018).

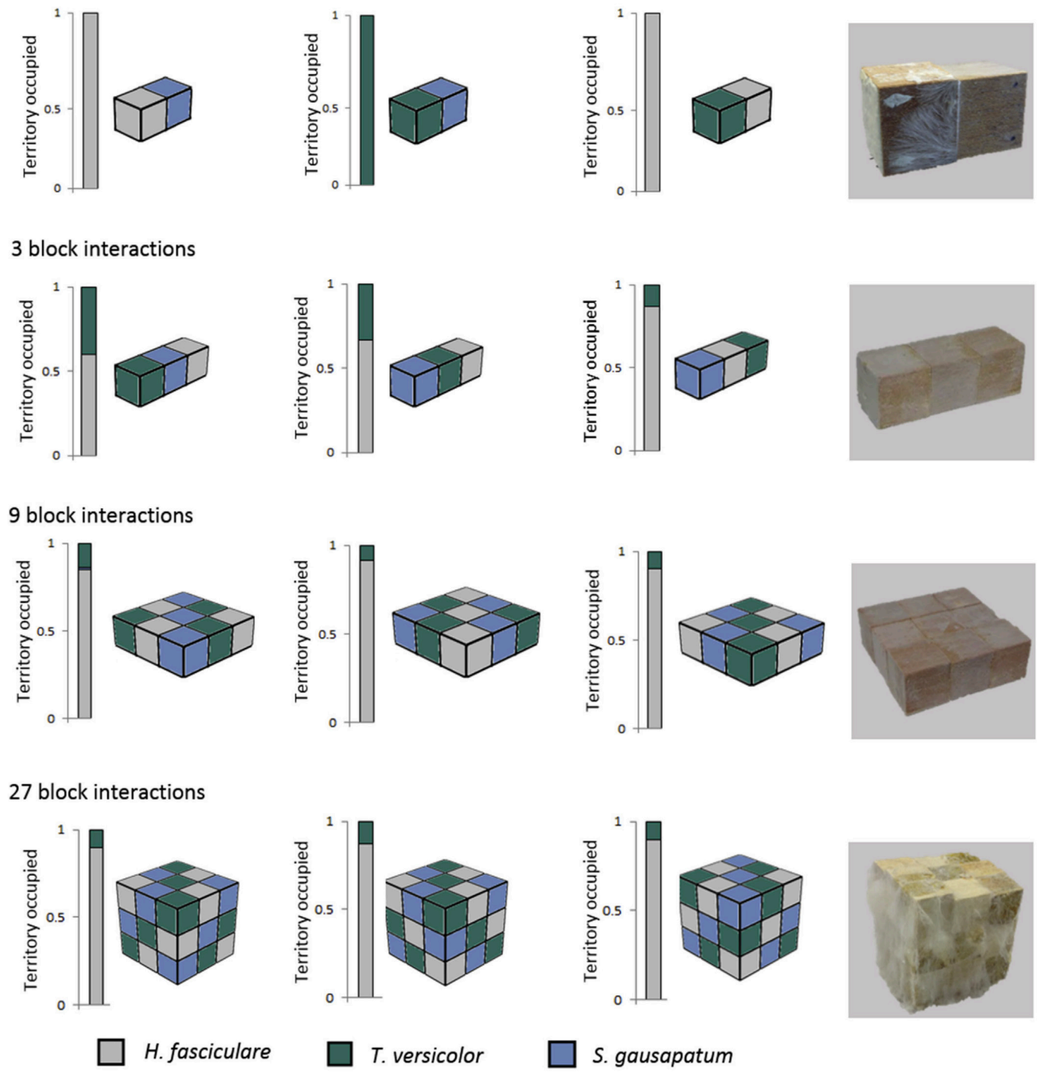


Figure 8

Multidimensional fungal interactions from the Fungus Wars study (Hiscox, O'Leary, and Boddy 2018).

5. Methods

5.1. Preliminary experiments

The preliminary experiments, as material experiments, used mycelium as a living material probe to explore the concept of a new biofabrication approach that allows making-through-growing. The aim of these mycelium experiments was to investigate the possibility of influencing mycelium growth by altering nutrients as a chemical environmental variable. The idea was to locally influence the aerial mycelium's development through nutrients, potentially enabling the creation of various forms with pure mycelium blocks.

Since no prior experiments had been conducted to influence the growth of aerial mycelium, and there was no established protocol, the primary objectives were to gain insights into setting up an experimental environment. This involved understanding key aspects such as; (i) inoculating mycelium spawn in agar, (ii) determining the required amount of agar, (iii) selecting appropriate ingredients for the agar media, and (iv) exploring whether nutrients could act as a negative factor in degrowth. In addition, these preliminary experiments were my first experience experimenting with materials in a lab environment, as a biodesigner. Since these were the first stages, it was essential for me to understand basic lab processes and methods to establish a basis for further research.

In preparation for the main experiment, a series of four preliminary tests were conducted to gain insights into the material and to determine the parameters for the main experiment. These preliminary tests also served as preliminary material experiments, allowing the designer to explore and interact with the material. This interaction helped to understand how to best structure, measure, document, and establish the scientific methods for setting up the main experiments.

Before running these experiments, an experiment plan, a Biological Control of Substances Hazardous to Health (BioCOSHH) assessment and a risk assessment form[2] were completed. These forms helped to identify any potential hazards and risks associated with the experiment and to ensure that appropriate measures were in place to minimise or control these risks. It further helped to ensure that the experiments were conducted responsibly.

[2] *Check appendix.*

The nutrient media used for all the preliminary tests and the main experiment was autoclaved at 121^{0}C for 15 minutes before the inoculation. Oyster mushroom (*Pleurotus ostreatus*) spawn, provided by GroCycle UK, was used as the strain. All the samples were inoculated and kept at room temperature during the growth.

I. Seeding layout pre-experiment and results

Before starting the nutrient deposition, it was necessary to understand the most effective way to inoculate the mycelium spawn on an agar plate. This involved determining the optimal positioning/layout of the spawn to maximise surface coverage and, in turn, promote efficient growth. The outcome of this test provided insights into seed positioning, which would be crucial for optimising growth in future experiments. During this test, all other factors remained

Table 2

The results of the seeding layout experiment.

Settling	Layout	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
		Surface coverage area (mm^2)					
Single seeds	[SS] Above	5	21	30	72	131	142
	[SS] Half buried	8	28	65	100	135	144
	[SS] Fully buried	3	15	36	85	130	144
	[SS] Floating	2	9	19	67	106	140
Groups of 4 seeds	[IN4] Above	3	9	21	47	69	98
	[IN4] Half buried	3	18	42	57	60	105
	[IN4] Fully buried	1	6	23	48	67	94
Groups of 6 seeds	[IN6] Above	3	10	17	38	53	83
	[IN6] Half buried	4	12	23	36	54	85
	[IN6] Fully buried	5	9	21	32	45	73
Groups of 12 seeds	[IN12] Above	2	5	13	22	31	51
	[IN12] Half buried	6	12	17	28	35	72
	[IN12] Fully buried	2	5	16	26	35	61
	[IN12] Floating	8	13	24	36	50	88

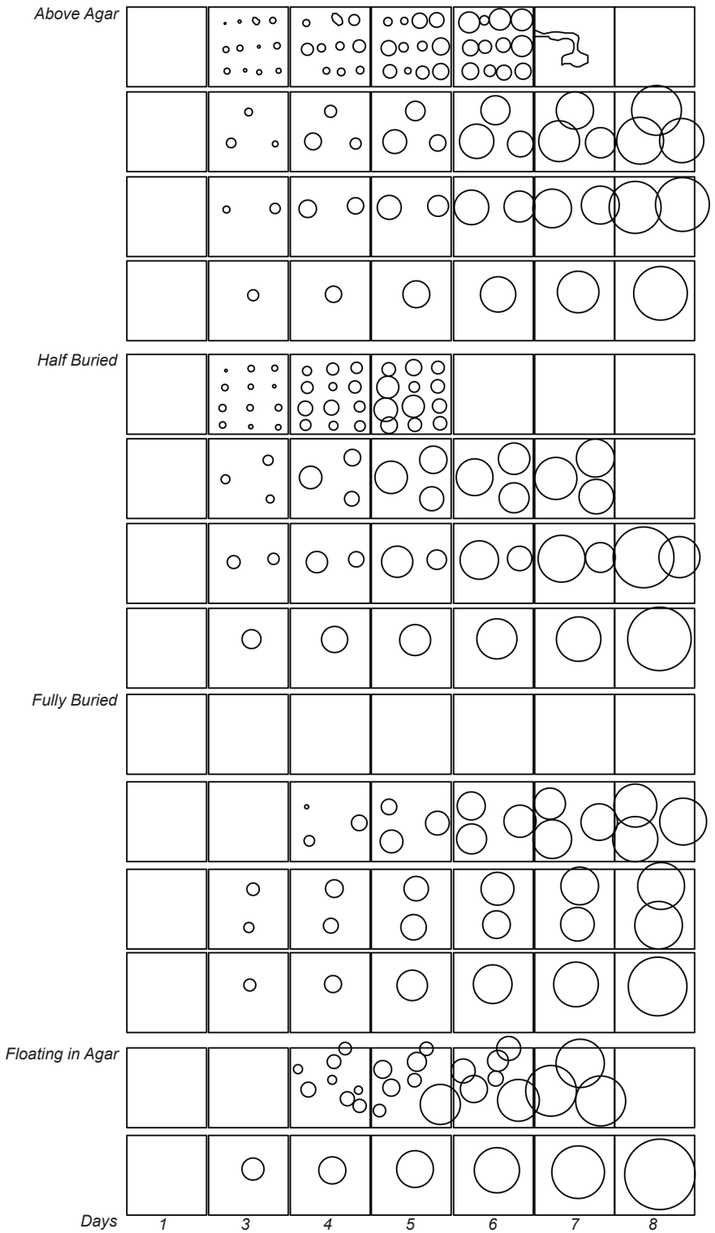


Figure 9

AutoCAD drawings of the petri dishes between Day 1 and Day 8.

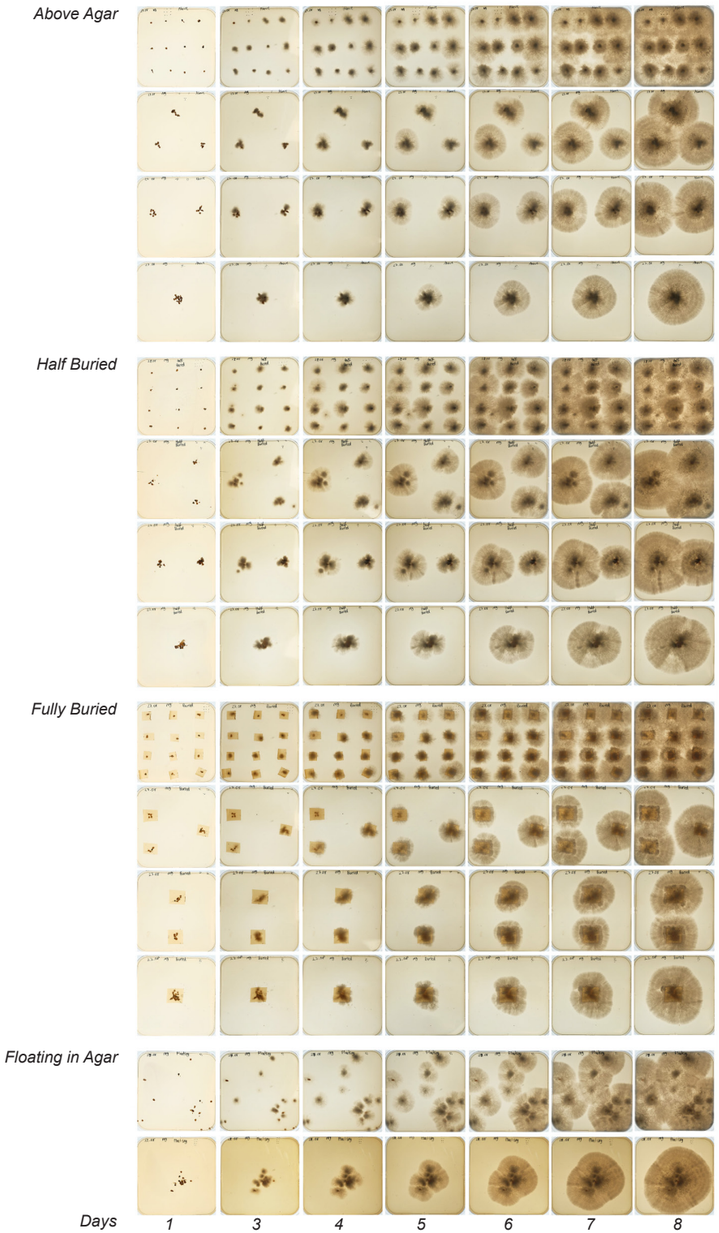


Figure 10

Photographs of the petri dish illustrating the seeding layout experiment between Day 1 and Day 8.

consistent, including the agar type (composed of 100g water, 4g malt extract, 2g dextrose, 0.16g yeast extract, and 2g agar), the quantity of mushroom spawn (12 seeds), and the petri dish size (10x10 mm). The only variable under investigation was the seeding method.

The first variable examined was the depth of seeds in the agar plates. This included seeds positioned above the agar, half-buried in the agar, fully buried in the agar, and seeds floating in the agar. The second variable involved the clustering of the seeds, with arrangements of twelve, six, four, and one seed. This resulted in a total of fourteen different seeding methods, as illustrated.

At the end of the eight days, the test was finalised, as there were no obvious changes in growth after day eight. Throughout the test, photographs were taken every other day using a Fuji film X-T2 camera equipped with an 80-mm lens. Photography helped to document and analyse the overall expansion and growth of mycelium. The surface expansion of mycelium was calculated by measuring from the photos. Photos were imported to AutoCAD, Figure 9, and adjusted to a 1/1 scale, allowing for digital measurement of the mycelium's expansion over a surface area.

The results showed that single seeding in a half-buried and fully buried layout gave the best mycelial surface coverage over the agar plate, as seen in Table 2 and Figure 10. For all the upcoming experiments, this single seeding, half and fully buried in agar layout, was used.

II. Agar amount pre-experiment and results

Once the positioning of the mycelium spawn in the agar was determined, the next question revolved around whether the quantity of agar affected the extent of growth in both the short and long term. Seeing the influence of the agar amount was important for gaining insights into the effects of nutrient deposition.

Mycelium was inoculated into 300-ml glass jars filled with different amounts of agar. To ensure even distribution of the mycelium spawn on the agar in these jars, an acrylic grid with 5x5 mm holes was prepared. The seeds were placed in the middle of each grid, resulting in uniform distribution, as seen in Figure 11.

For measuring the mycelium thickness within the jars, an acrylic plate with six holes was prepared using a laser cutter. This plate was positioned on top of the jars, and fret saw blades were inserted through these holes to ensure measurements were taken from the same points consistently. Mycelium thickness was determined by calculating the average of these measurements across the six points. The volumetric growth was measured by multiplying the surface area and the thickness of mycelium. To measure the thickness, thin ethanol-resistant fret saw blades (16 TPI) were used, as seen in Figure 11. These blades could be easily inserted into the agar. To mark the highest point in the agar covered by mycelium, the blades were first wiped with 70% ethanol and then dipped into the agar. Finally, the marked point was measured with a ruler.

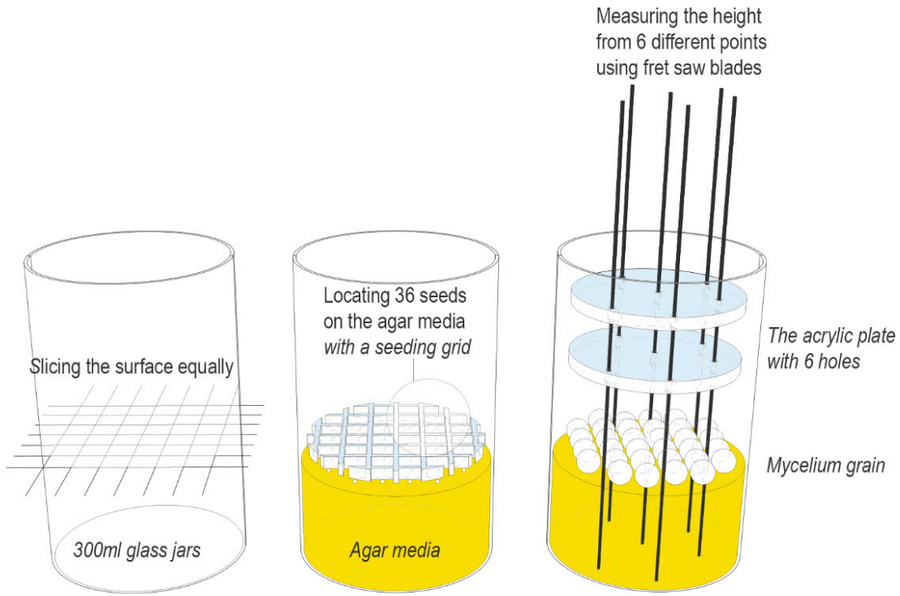


Figure 11

A photo and diagram of the seeding and measuring technique.

Table 3

The results of the agar amount experiment.

Agar Amount	**20 ml**			**40 ml**			**80 ml**			**160ml**		
Time (day)	8 days	16 days	30 days	8 days	16 days	30 days	8 days	16 days	30 days	8 days	16 days	30 days
Volume (cm3)	4.95	6.05	7.98	4.13	6.88	7.98	5.23	8.25	8.53	4.13	8.53	9.08
Error of the mean	0.55	0.55	0.27	0.28	1.93	0.27	0.28	2.75	0.28	0.28	0.28	0.27

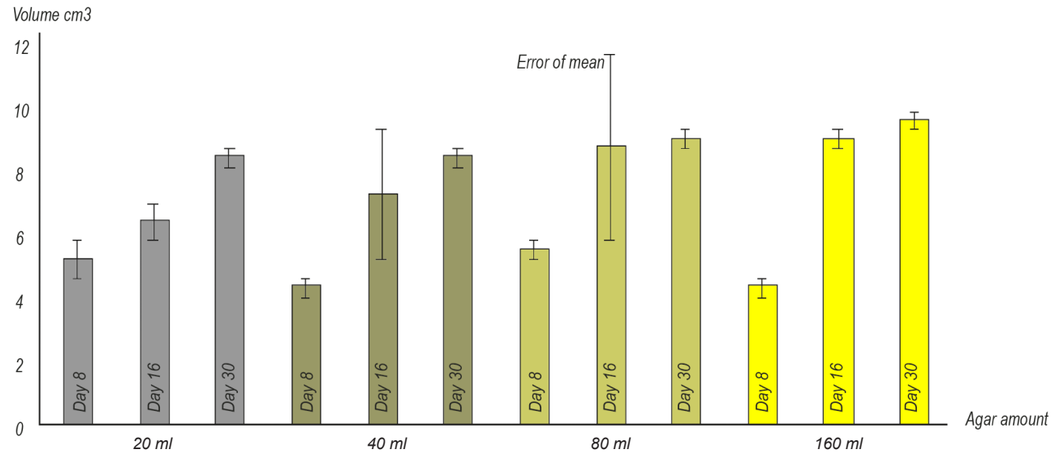


Figure 12

The volumetric growth of agar amounts experiment in numerical and column chart format.

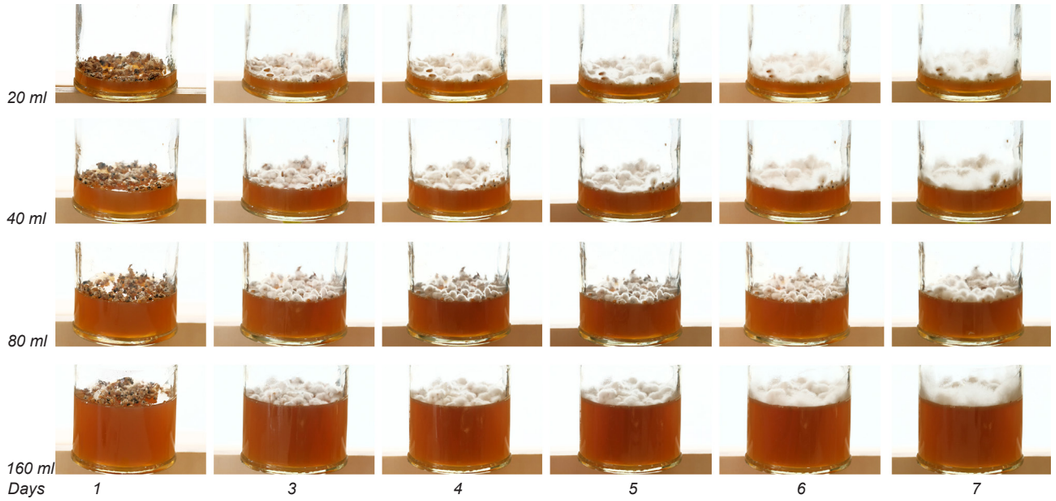


Figure 13

Photographs documenting the agar amounts experiment from Day 1 to Day 7.

The agar media (consisting of 4g of malt extract, 2g of dextrose, 0,16g of yeast extract, and 2g of agar for 100g of water) remained consistent, but the volume was adjusted to 20 ml, 40 ml, 80 ml, and 160 ml. To start the experiment, 36 oyster mushroom seeds were inoculated into a 300-ml glass jar using the acrylic grid after thoroughly cleaning the jar with 70% ethanol. The test ran for 30 days. The experiment was carried out in triplicate, allowing for the determination of the average mycelium thickness formed under each specific condition. Additionally, the standard error of the mean was calculated using an Excel spreadsheet to assess the deviation between the samples.

The lids of the jars were closed after the inoculation. However, the difference in the level of air due to jar lids being closed was not accounted for. Therefore, the results might be skewed. This possible effect could be eliminated, and the air could be equalised by replacing the lids with a cheesecloth.

The results showed that during the first eight days, there was no significant difference in mycelium thickness among the various agar jars. However, after two weeks, the volume of agar had a positive impact on promoting growth. The thickest mycelium layer was observed in the 160-ml agar jar (Figure 12). From this experiment, it can be concluded that volumetric mycelium growth increases in the long term with with larger agar volumes, as shown in Table 3 and Figure 13.

III. Agar culture pre-experiment and results

The agar culture test aimed to reach the best volumetric growth through the change in the nutrient medium in order to understand the oyster mushrooms' preferred habitat. This information would be valuable in cultivating mycelium in specific three-dimensional forms through nutrient deposition. Thirteen different nutrient combinations, consisting of dextrose, fructose, malt extract, and yeast extract, were used, as outlined in Table 4. The same seeding and measuring grids were used to set the experiments and measure the outcomes. The experiment was conducted in triplicate, and the standard error of the mean was calculated using Excel Spreadsheet. The test was finalised at the end of the eighth day.

The results indicated that the combination of malt extract, yeast extract, and dextrose in Media-11 produced the most favourable mycelial growth for oyster mushrooms, as shown in Table 5 and Figure 14. According to the results:

- Whilst malt extract was enough for growth on its own, dextrose alone was not sufficient for the mycelium extension.
- Fructose and dextrose had a similar impact on the mycelium growth.
- The addition of yeast extract to the mixture enhanced growth, as observed in Media-9.
- Media combinations containing malt extract and dextrose in equal proportions did not significantly contribute to growth.

Table 4

The ingredients of thirteen different agar media.

M-1 [2.1%]	M-2 [4%]	M-3 [0.5%]	M-4 [1.3%]	M-5 [6%]	M-6 [6%]	M-7 [4%]	M-8 [3.2%]	M-9 [6%]	M-10 [6.4%]	M-11 [6.4%]	M-12 [4.3%]	M-13 [6%]
100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water	100ml water
	2gr malt extract	0.1gr malt extract	0.3gr malt extract	6gr malt extract	2gr malt extract	4gr malt extract	1gr malt extract	4gr malt extract	4gr malt extract	2gr malt extract	4gr malt extract	4gr malt extract
2 gr dex-trose	2 gr dex-trose	0.3gr dex-trose	0.6gr dex-trose		4gr dex-trose		2gr dex-trose	2gr dex-trose	2gr dex-trose	4gr dex-trose		2gr fruc-tose
0.16gr yeast extract		0.04gr yeast extract	0.08gr yeast extract				0.24gr yeast extract		0.48gr yeast extract	0.48gr yeast extract	0.32gr yeast extract	
2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar	2 gr agar

Table 5

The results of the agar culture experiment.

	M-1 [2.1%]	M-2 [4%]	M-3 [0.5%]	M-4 [1.3%]	M-5 [6%]	M-6 [6%]	M-7 [4%]	M-8 [3.2%]	M-9 [6%]	M-10 [6.4%]	M-11 [6.4%]	M-12 [4.3%]	M-13 [6%]
Vol (cm3)	14.55 ±0.6	15.47 ±1.6	11.42 ±0.2	18.24 ±1.3	15.66 ±2.7	15.66 ±0.2	12.53 ±0.9	22.11 ±0.9	17.13 ±1.6	23.40 ±2	26.34 ±2.7	16.03 ±0.3	21.37 ±0.9

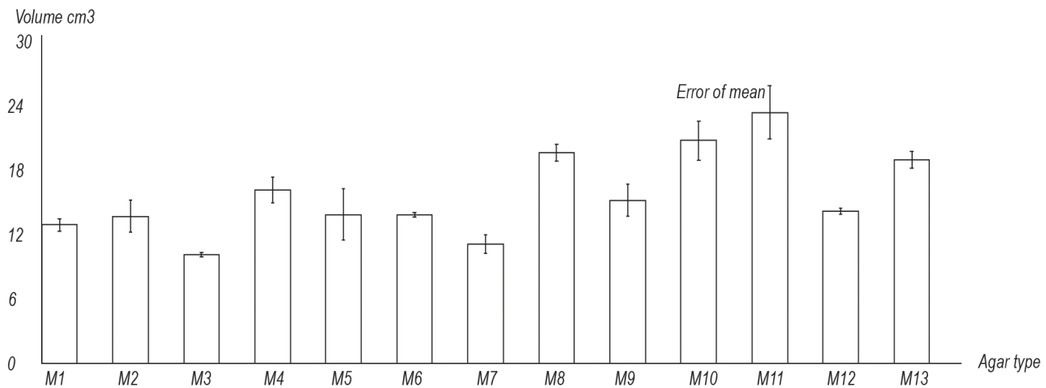


Figure 14

Comparison of volumetric growth for agar amount experiment.

Figure 15

Photographs of mycelium growth in thirteen different agar cultures on Days 1,3 and 8.

- Malt extract had a more substantial impact on growth compared to the dextrose-only mixtures, as there was still mycelial growth in the malt-only media.
- In the dextrose-only media, the hyphae organisation was very delicate, as seen in Figure 15.
- A 6% nutrient concentration with malt, dextrose, and yeast gave the best growth results.

IV. Growth halting pre-experiment and results

The growth halting test aimed to explore methods for intentionally inhibiting mycelium growth in agar culture. This exploration helped to understand how to negatively impact the organism, either by promoting degrowth or inducing deception that implies contraction. Understanding how to intentionally inhibit mycelium growth could be useful for conducting controlled experiments, for more precise and targeted morphological outputs.

Stressors, such as acetic acid, salt, white vinegar, and sodium bicarbonate, were used to induce stress in the mycelium (Table 6). To test growth halting, the mycelium spawn was inoculated into a divided petri dish. One half was provided with a stress-inducing substance, and the other half used Media-11. These were allowed to grow together for 14 days, as seen in Figure 16.

No growth was observed in any of the media, as seen in Figure 17. This result suggests that specific interventions using stress sources can be employed to sculpt mycelium biomass or inhibit its development at specific stages. While salt does not cause mycelium to contract, it is unclear whether sodium bicarbonate and vinegar have the same effect.

The four preliminary experiments helped me gain confidence and increase familiarity with working with mycelium. With each preliminary experiment, a better understanding of how to set and move on the main experiments developed. These tests helped in understanding:

Table 6

The ingredients of growth halting media.

Media K-1	K-2	K-3
100ml water	100ml water	100ml water
4 gr salt	4 gr sodium bicarbonate	6ml white vinegar
6 gr malt extract	6 gr malt extract	6 gr malt extract
Oyster mushroom seeds	Oyster mushroom seeds	Oyster mushroom seeds
2 gr agar	2 gr agar	2 gr agar

Figure 16

Diagram of the setting for the growth halting experiment.

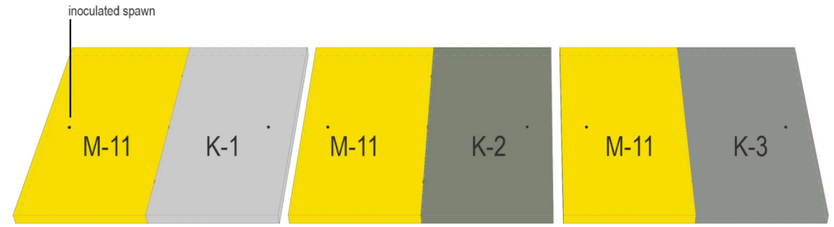
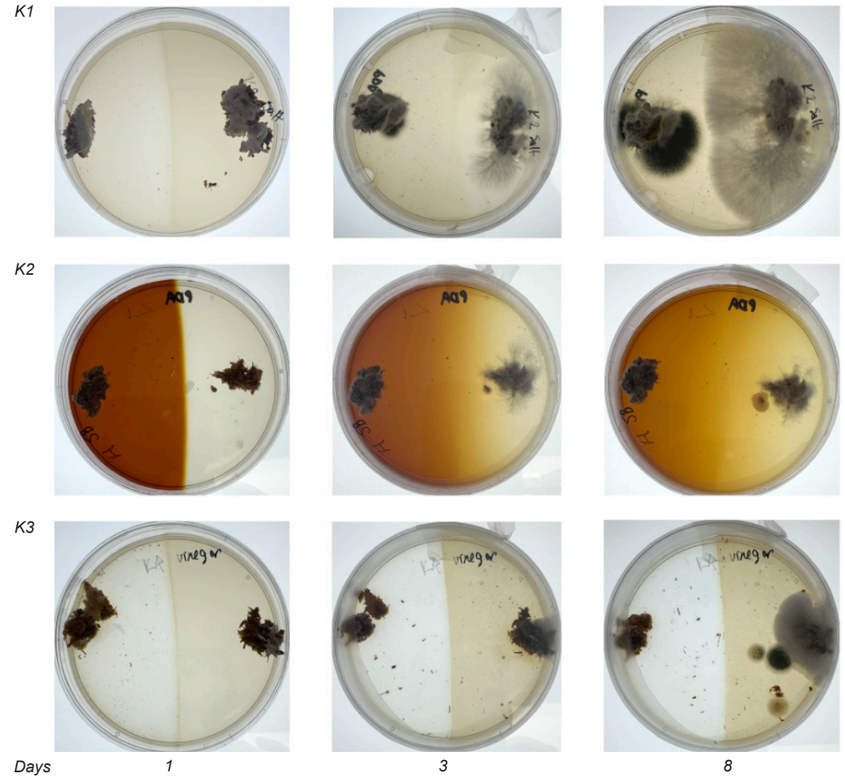


Figure 17

Photographs of mycelium growth on agar plate filled with half M-11 and half K-1,2 and 3 media.



1. Inoculation techniques for the spawn,
2. The effect of the agar amount on the growth,
3. The effect of agar type on the mycelium growth,
4. Strategies to block mycelium growth.

The main experiment was designed in light of these findings from the preliminary tests. A system that combines all the findings from the preliminary tests was designed to guide the growth of mycelium.

5.2. The Main Experimental Design

Considering the decisions made from the preliminary tests, the main experiment was designed and conducted. It aimed to merge the information obtained from the previous four preliminary tests. The goal was to generate nutritionally heterogeneous environments by using different paths with high to low nutrient status for the mycelium to follow, grow, and adopt its morphology.

A waffle structure with 25 cubicles (2x2 cm) was built from acrylic (using a laser cutter) and plastic (using a vacuum former). Then, each cubicle was filled with three different mediums derived from the agar culture (Media-6 and Media-11) and growth halting experiment (Media-K.3), as seen in Figure 18. The patterns were randomly generated, and only two of them were used in the experiment. In each cubicle, five mycelium seeds/spawns were carefully inoculated using tweezers within a laminar flow cabinet. Following inoculation, the waffle structure was sealed with a clean film and the mycelium was allowed to grow for 14 days to maximise its growth potential. The underlying assumption was that mycelium would exhibit more robust growth in its preferred mediums and comparatively less growth in less favoured ones.

The objective was to generate varying aerial mycelium thicknesses and formations using this method, an aspect that could play a key role in shaping the form of the growth. The experiment also aimed to explore the sensitivity of organisms in this stage of development.

6. Results

The results of this experiment revealed that the fungal community acted as expected in laboratory settings. Aerial mycelium thickness and density displayed an increase in the cubicles with the most favourable media, and not an increase in those filled with the least favourable media, as seen in Figure 19.

As seen in Figure 20, hyphae growing in various chemical environments demonstrated distinct amounts of growth; however, there was no apparent morphological difference on a larger scale. This shows that the mycelium demonstrated developmental plasticity at a microscopic level, but these changes did not affect the overall form on a macro scale.

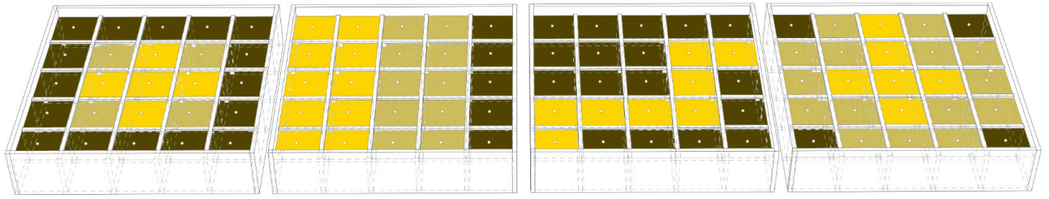
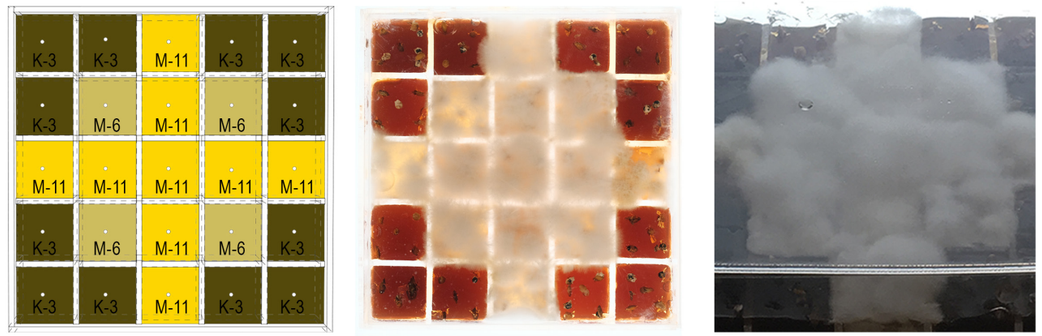


Figure 18

Diagram of the setting for the growth halting experiment.



The setting *Day 8*

The setting *Day 8*

Figure 19

The setting of the tessellated agar (left), the mycelium growth on Day 8 (centre), zoomed images of the experiment (right).

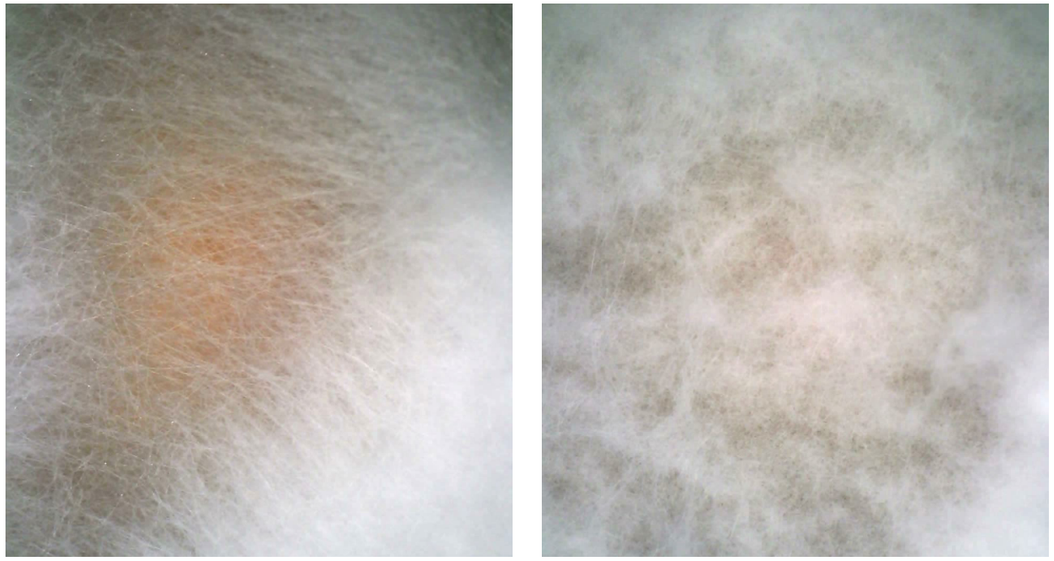


Figure 20

Microscopic images of aerial mycelium growing in M-11 (right) and M-6 (left) (Dino-Lite digital microscope at 70X magnification).

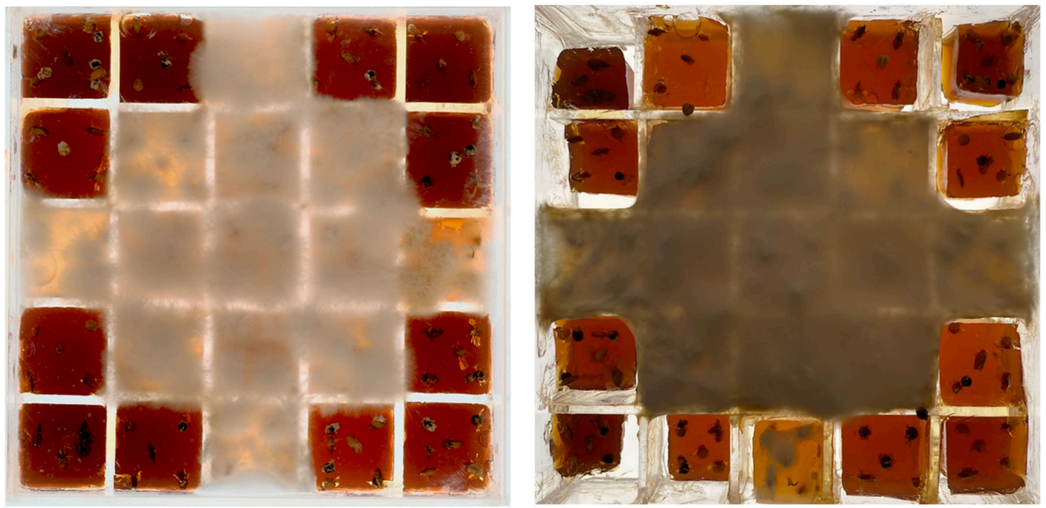


Figure 21

Photography of inoculated seeds on Day 8 (left), inoculated seeds on Day 40 (right).

After two weeks, the growth stopped, and the mycelium began to wilt away and dry out due to decreased nutritional values, as seen in Figure 21. To prevent this decline, an irrigation system was required. Instead of continuing to feed the mycelium, the mycelium experiments were stopped at this point, as the aerial mycelium did not demonstrate self-assembled anatomy on a macro scale without the need for scaffolds or control structures, such as moulds.

7. Discussions

After the four preliminary tests and the main experiment had been conducted, it was concluded that controlling the growth by continuously feeding mycelium from specific points can be used as a (limited) method to guide the overall mycelium growth. Each feeding cubicle can be imagined as pixels generating the whole geometry. However, nutrition needs to be pumped to the individual cubicles for further growth, as seen in Figure 22 (left). By receiving food with more or less favourable nutrients, the mycelium will start to rise or not rise from the plane. Mycelium in the more fed regions will grow more abundantly, helping to form the overall geometry. Although mycelium does respond to changes in its chemical environment by increasing growth during this process, this method cannot be used as a biofabrication strategy that allows making-through-growing.

After a time, the increasing number of cells in favourable nutritious cubicles may reach a point where further growth is not possible due to overcrowding. Additionally, the seeds in the cubicles with the less favourable nutrients would exhibit further growth due to the space around them. At that point, there will be a need to have a scaffold for the mycelium to cling to, as seen in Figure 22 (right). Otherwise, the system would reach a point where there is a flip in the growth, and favourable cells would provide worse growth results due to overcrowding.

Moreover, a limitation of this mechanism, when considering its application in creative practices, is that it only nourishes mycelium in a single direction. This restricts the potential to create more complex structures; as aerial mycelium primarily grows upwards from the horizontal plane, limiting variations in the vertical or sectional plane. As a result, it was concluded that while the tessellated agar spot method from Ritz and Crawford (Ritz and Crawford 1997) allows designers to manipulate the mycelium's density and surface area, it provides little flexibility in altering the overall morphology.

In terms of the automatization of the system (to slide the fabrication towards the digital side from the manual), the cubicles could be fed with a digitally controlled feeding mechanism in a tessellated agar system. However, such a mechanism will not be the smartest way of taking advantage of digital tools. Pumping nutrient media to the cubicles, which does not require intelligence, can be done by the designer or a mechanical system.

Mycelium experiments have also contributed significantly to the comprehension of various states of developmental plasticity. They have showed that the reason pumping nutrients cannot be used as a 'making-through-growing' strategy. The hyphal morphology, when fed with different nutrients, will exhibit slight differences on a microscopic scale. However, dur-

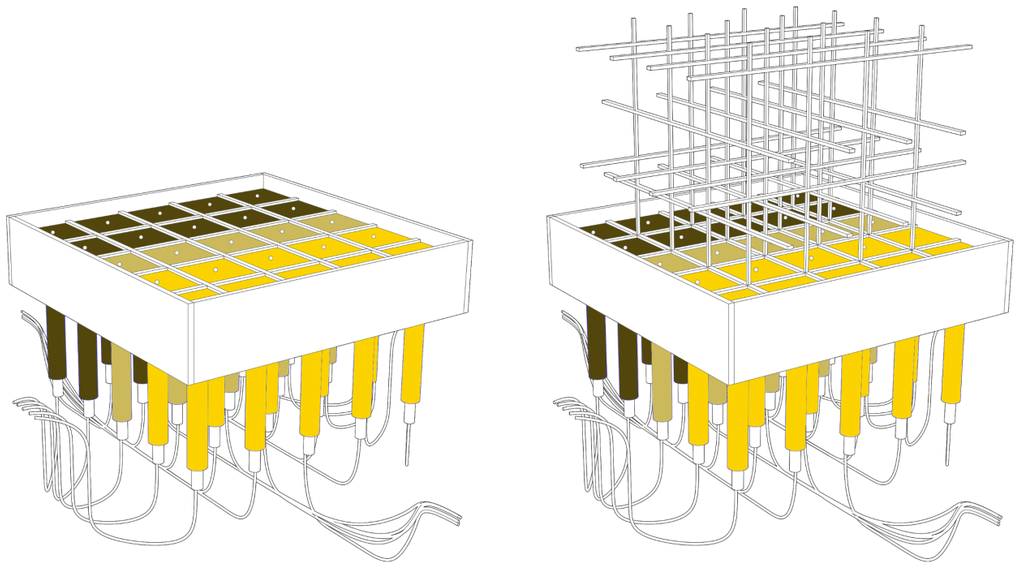


Figure 22

Diagram of the proposed setting to generate patterns with mycelium growth; Cubicles with nutrition pumps (left) and cubicles with nutrition pumps and scaffold (right).

ing this process, hyphal filaments will intertwine, and their developmental plasticity will not be visible to the human eye. This means that mycelium cannot give form as a result of its morphological adaptability.

The distinct life stages of the organism (such as fungus in mycelium and mushroom form), as well as different parts of the same organism (such as roots and fruiting bodies or leaves and roots of a plant), may exhibit varying levels of morphological change due to developmental plasticity. While mycelium in the experiments did not exhibit many phenotypic changes on a macro scale, it remains accurate to say that the fungus demonstrates developmental plasticity. This is evident in its ability to adopt diverse life forms, serving as both hyphae—a branching structure—and mushrooms.

As mentioned in the introduction, plasticity in philosophy is defined as “the capacity to receive and give form” (Zukauskaite 2023). According to this definition, it can be argued that mycelium is good in form-taking, but not in form-giving. Mycelium receives a specific form by consistently following the food source, consuming nutrients, and adapting to the provided form. It has the ability to fill the negative space of a provided mould with minimal autonomous control over its shape. However, mycelium does not exhibit the same proficiency in form-giving, especially on a macro scale. Therefore, rather than characterising it as less plastic, it is more accurate to describe its capabilities in terms of form-giving versus form-taking.

Based on the difference in form-taking versus form-giving characteristics, when mycelium turns into a mushroom, it becomes a form-giver, enabling significant changes in morphology based on environmental cues. Morphologically, hyphae and mushrooms represent distinct developmental entities due to their constant responsiveness to the environment, despite sharing the same genes. Consequently, after confronting the limitations of working with mycelium and conceptualising how to cultivate it in the desired geometry, the decision was made to focus on the fruiting bodies. Fruiting bodies are also formed by the filamentous hyphae (mycelium) and have an intrinsically more varied morphology. The transformation from mycelium to a large, reproductive, and multicellular structure, such as mushrooms, demonstrates developmental complexity and tissue differentiation. Unlike mycelium, mushrooms are self-organising and possess a structure that allows them to stand independently.

8. Conclusion

This chapter covered the concept of plasticity and its implication for biological developmental processes. It illustrated these ideas through mycelium experiments, which served as the realisation of conceptual thought experiments through physical design and craft processes. It suggested that developmental plasticity can potentially enable a parametric approach through non-physical intervention, serving as a biofabrication method. As Dade-Robertson discusses, indirect methods of affecting a living material through environmental parameters use “nature’s own agencies” without human imposition through “forcible constraints” (Dade-Robertson 2021). However, the experiments in this chapter, conducted with mycelium, showed that fabricating it with minimal intervention through its adaptive plasticity has

challenges.

Therefore, it can be concluded that mycelium exhibited the capability to adapt its form at a microscopic level in response to variations in nutrient levels. The hypha adjusted its density and thickness in response to diverse nutrient sources. However, these changes were only visible at a micro scale and did not lead to significant alterations in the overall 3D form at a macro scale. The microscopic changes did not induce noticeable alterations in overall morphology. The observed developmental plasticity in mycelium during this series of experiments was relatively limited, as the chemical environmental parameters did not trigger the organism's transformation into other life stages, such as the growth of hyphal knots or fruiting bodies. This implied that fabricating some organisms based on their morphological adaptivity is challenging. Consequently, further experiments have shifted their focus to mushrooms, which inherently have more morphological complexity, demonstrating visible adaptations to their environment.

The next chapter focuses on the fungal fruiting bodies and further questions whether their developmental plasticity demonstrates linear behaviour when influenced by the designer. Since biological processes often exhibit non-linear behaviour, the same effect does not always cause the same results (Carpo 2013). Even small changes in environmental conditions can lead to tipping points and it can result in various developmental outcomes that are not easy to attribute to single or small sets of parameters. Furthermore, the same set of conditions does not always produce identical results; an organism growing under identical environmental circumstances may exhibit different morphologies. Biological systems are also subject to noise, and exhibit emergence where outcomes cannot be easily reduced to a single cause or an individual change. This biological complexity, therefore, challenges a parametric approach. Therefore, the next chapter will continue exploring the question of whether fruiting bodies, as adaptive biological systems, exhibit parametric responses.

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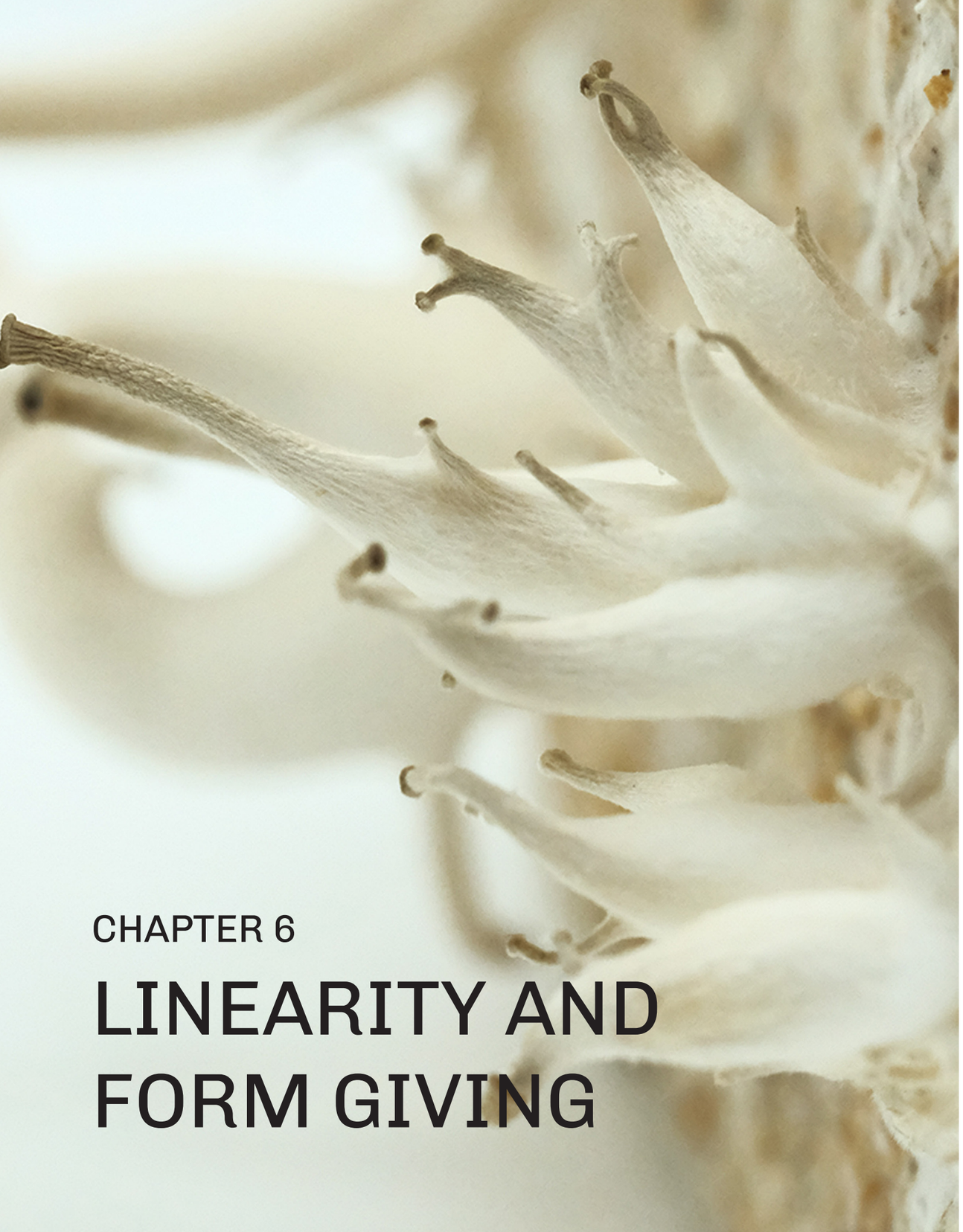


CHAPTER 6

LINEARITY AND FORM GIVING

1. Introduction

Chapter 5 demonstrated that mycelium lacks form-giving abilities on macroscales, which could be attributed to material interactions during its growth through adaptive plasticity. Therefore, this chapter shifts its focus to fungal fruiting bodies, as they present a more intriguing subject for study due to the complex relationship between macro form and environmental parameters. The primary goal of this chapter remains the same: the development of a biofabrication method and tool to guide the growth of living materials through their developmental plasticity.

To achieve this objective, the chapter introduces the concept of biological-parametric design as a fabrication strategy through design experiments. These design experiments investigate the relationship between environmental parameters and fungal fruiting body morphology. The initial step involves addressing the question: What are the most critical parameters influencing fungal (fruiting body) morphology, and how do these parameters relate to each other? Answering this question helps to distinguish variables in terms of their cause and effect, as well as to explore the interrelation between input parameters and output morphologies.

After addressing the parameters, systematic and control-based experiments focusing on fruiting bodies will be employed. A digitally controlled environment, a growth chamber, will be designed and built to serve as a parametric tool for biofabrication. To design a digitally controlled growth chamber, its appearance, functionality, and impact on mushroom development will be explored. Given their digital control, these chambers can offer a high degree of control, allowing for reliable, precise, and repeatable experiments.

The mushrooms produced from these experiments help to demonstrate a broader concept, indicating a new type of parametric design for living material. The exploration of environmental parameters influencing diverse fruiting body morphologies raises questions about the nature of these input-output relationships—whether they are ‘linear’, ‘nonlinear,’ ‘correlational,’ or ‘predictable.’

Chapter 6 *is reworked from the journal D. Ozkan, M. Dade-Robertson, R. Morrow, M. Zhang, Are mushrooms parametric? in Biomimetics Special Issue, Fungal architectures, 2022, and from the conference proceeding D. Ozkan, M. Dade-Robertson, R. Morrow, M. Zhang, Designing A Living Material Through Bio-Digital Fabrication, in eCAADe Towards a New Configurable Architecture, 2021.*

2. Linearity and Correlation

Linearity, as a mathematical property, refers to the way in which a system responds to diverse inputs. In a linear system, outputs are directly proportional to inputs (“Variables, Functions and Equations,” n.d.). In other words, if the input variables change, the output variable will change by a fixed multiple (or proportionally). A mathematically linear relationship is only characterised by a straight line. Linear systems are relatively simple and easy to understand because the relationship between the input and the output is straightforward and predictable.

On the other hand, a nonlinear system is one where the output does not maintain a direct proportionality to the input (“Variables, Functions and Equations,” n.d.). The relationship between the input and output becomes more complex and less predictable. Such systems can exhibit a wider range of behaviours, including exponential growth, oscillation, and chaotic behaviour (Figure 1). Nonlinear systems are often more difficult to understand and predict compared to linear systems, primarily because the relationship between input and output is more intricate and less predictable.

Although the concept of linearity is straightforward, according to DeLanda, it can be a subset of a variety of nonlinear possibilities (2015). In many cases, a linear model can be seen as an approximation of a more general nonlinear model. This is primarily due to the simplicity and comprehensibility of linear models, making them a commonly used starting point for more complex analyses. It is important to recognise that linear models only offer an approximation of the broader nonlinear scenario, and they may not accurately represent the system’s behaviour under all circumstances (DeLanda 2015).

DeLanda takes the ‘Same cause, Same effect, always’ formula of linear relationship, and questions its validity or problematic nature in certain cases (DeLanda 2015). He argues that the same can be interpreted as intensity (‘Same intensity of cause, Same intensity of effect, always’). For example, Hooke’s law assumes linear behaviour, presenting a proportional response to a specified cause, such as load and material deformation (DeLanda 2015). However, some materials, like organic tissue or rubber, display exponential or quadratic curves when a load is applied. A balloon, for example, can inflate to a certain point, but explode, responding differently to the load beyond that threshold. From a mathematical perspective, the behaviour of these materials is nonlinear because the terms “linear” and “nonlinear” refer to whether the relationship between variables can be described as a straight line. However, it is important to note that “linear” and “nonlinear” are not a dichotomy, and various correlations between variables can generate nonlinear (S- or J-shaped) curves.

DeLanda identifies another issue with the ‘Same cause, Same effect, always’ formula, specifically with the inclusion of the term “always”. When transitioning from an individual entity to a population, causality becomes statistical. For example, although smoking increases the risk of cancer, it cannot be asserted that every smoker will inevitably develop cancer; rather, smokers have a probability of developing cancer. Similarly, in mushroom experiments, an increase in CO_2 may result in small mushroom caps, yet it cannot be definitively stated that

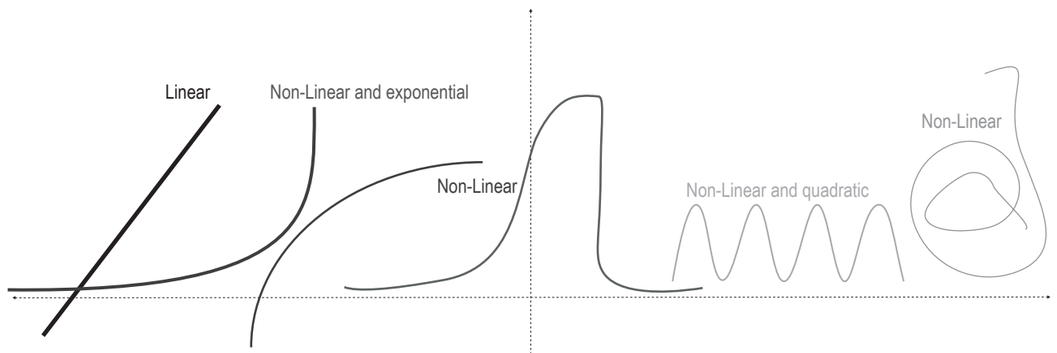


Figure 1

Different graphs of linearity and nonlinearity defined as mathematical functions.

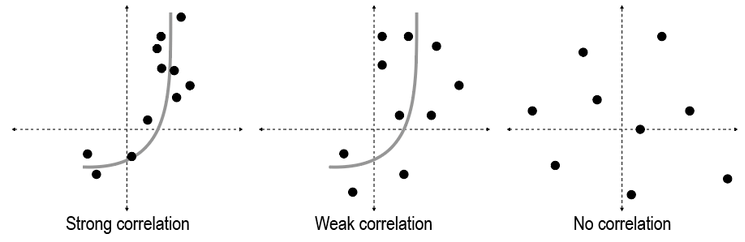


Figure 2

Diagram of three types of correlation.

every individual mushroom will exhibit a small cap in high CO_2 conditions. It is more accurate to describe this outcome as likely or probable. The slight chance of the mushrooms not developing small caps makes them a nonlinear system.

Linearity is used for simplification in the analysis and prediction of variable relationships due to its ease. However, it is important to note that many real-world relationships between variables are nonlinear and cannot be described by a straight line. Nonlinear relationships between variables can be more complex and difficult to predict, but often more accurate and detailed descriptions of real-world phenomena. They cover complexities and systems near equilibrium, such as exponential relationship. Exponential relationships effectively capture the complex behaviours that occur as the system approaches equilibrium. Overall, it is critical to consider both linear and nonlinear relationships when trying to understand and describe the real world. Linear relationships are often useful as a starting point, but nonlinear relationships may be necessary to fully grasp the system's complexity.

The design experiments in this chapter will be used to understand the relationship between morphology and environmental factors. If there is a correlation between input and output variables, it implies that the mushrooms can exhibit a degree of predictability in various parameters of growth and form. Correlation is a measure of the strength and direction of the relationship between two variables. It can be characterised as strong or weak, as seen in Figure 2 ("Types of Correlation," n.d.). Importantly, correlation does not necessarily imply causality but does provide information about the relationship between two variables. A strong correlation indicates a well-defined and predictable relationship, while a weak one suggests less predictability. When there is no discernible pattern between the variables, there is no correlation. When there is a relationship between variables, but the relationship is not linear (a straight line), there is a nonlinear strong or weak correlation ("Types of Correlation," n.d.).

Determining whether mushrooms demonstrate linearity, nonlinearity or correlational behaviour is essential for the development of a biofabrication method that facilitates material interaction based on specific parameters.

3. Fungal Fruiting Bodies

3.1. A General Overview of Mushrooms

As mentioned earlier, mushrooms were chosen as an ideal organism for this research due to the following reasons: rapid growth rate, totipotency, and high developmental plasticity. Totipotency refers to the capacity of a cell to divide and autonomously produce all the differentiated cells of an organism (Kaul and Ventikos 2015). This means that if even a small piece of mushroom tissue is transplanted onto a nutrient medium, it can initiate new growth and create a colony of branching cells (Watkinson, Boddy, and Money 2016). The transformation of a single tissue cell from filamentous hyphae (mycelium) into a large reproductive and multicellular structure, such as mushrooms, demonstrates developmental complexity and tissue differentiation.

The ability of fungal tissue to undergo a sophisticated developmental pathway makes mushrooms a suitable organisms to work with. It facilitates easy experimentation, as cells harvested from one experiment can be used as the basis for a new one. Totipotency provides simplicity by allowing the use of a single base cell, serving as raw material, without the need for additional organisms. This can lead to the formation of a hypha, mycelium, hyphal knot, and fruiting body formed by a stipe (stalk), supporting a pileus (cap), and spores during its lifetime.

The morphological diversity inherent in the growth process provides a broader range for the designer to intervene. Developmental plasticity allows a single cell to exhibit a spectrum of morphological changes, enabling mushrooms to adapt to their environment. There are 16,000 identified mushroom-forming species, differentiating themselves more obviously with their fruiting bodies (Watkinson, Boddy, and Money 2016). The formation of the mushrooms allows the most effective diffusion of spores. The umbrella shape of mushrooms comes from the upward development of stipes under the influence of light, whereas the gills that diffuse the spores develop downward due to gravity (Sakamoto 2018). Finally, the rapid visibility of morphological adaptations in mushrooms makes them suitable organisms to work with for the limited duration of this research.

3.2. Fruiting Body Growth

As mentioned in Chapter 5, the type of fungi used in this research belongs to the *Basidiomycota* group. The growth of *Basidiomycota* involves two main stages: the vegetative phase (hyphal growth) and the reproductive phase (mushroom growth) (Kues 2003). Phosphorus, sulphur, potassium, calcium, and magnesium are the elements and minerals that are necessary for the reproduction of any kind of fungi (Watkinson, Boddy, and Money 2016).

The growth process begins with the germination of male or female spores of *Basidiomycota* to form a branching hyphal colony. This branching of a single-sex mycelium encounters the opposite sex, and they fuse. When they fuse, they double their genetic information (Kues 2003). These doubled mycelium cells maintain their unique properties during cell division, ensuring the transportation of all genetic information (Watkinson, Boddy, and Money 2016).

The reproductive phase follows the creation of the mushroom (reproductive phase). The colony of mycelium undergoes significant changes through the expansion and inflation of pre-existing hyphae, forming hyphal knots while generating fruiting bodies. Normally, the hyphal filaments show positive autotropism by growing outward. However, during the formation of a fruiting body structure, they begin to grow inward, and display negative autotropism (Moore et al. 2008).

Knots serve as the starting points for visible fruiting bodies, which can be triggered by disturbances such as an injury, encounters at the edges, or changes in nutrient levels, temperature, or light exposure (Moore et al. 2008). Depending upon the species' phototropic requirements, progress may proceed with a regular light-dark cycle, leading to cellular differ-

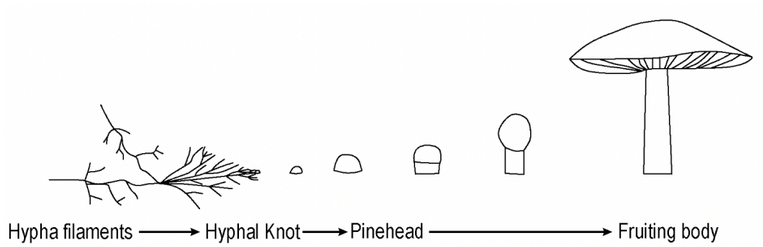


Figure 3
The developmental path of the fungal reproductive phase.

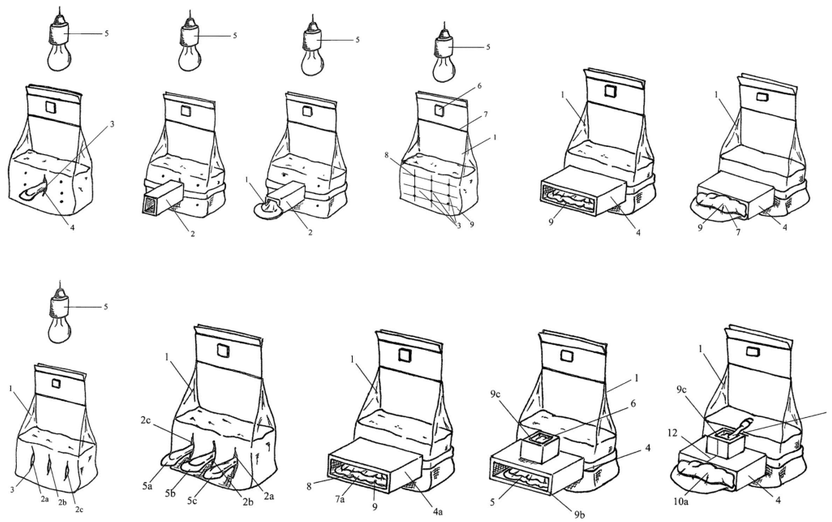


Figure 4
Method for producing rapidly renewable chitinous (the most abundant polymer in nature) material using fungal fruiting bodies and product made thereby (Bayer and McIntyre 2011).

entiation (Kues 2003). Under suitable conditions, they develop into both a pinhead and the fertile tissue of a fruiting body (Figure 3).

The formation of the stipe, cap, and gill cells occurs during the formation of the fruiting body, giving the mushroom its characteristic appearance (Watkinson, Boddy, and Money 2016). Spores are discharged from the surface of the gills. Hydrostatic pressure, airflow, raindrops, and surface vibrations all contribute to spore discharge (Watkinson, Boddy, and Money 2016). Gills increase their surface area through folding, allowing the production of more spores (Watkinson, Boddy, and Money 2016).

A basidiomycete mushroom can shed 30 billion spores per day (Watkinson, Boddy, and Money 2016). Although spore dissemination is influenced by the environment, it is observable that a reduction of nutrients in the area results in a more fragile fruiting body, releasing more spores over a wider radial area (Watkinson, Boddy, and Money 2016). This can be interpreted as the fungus's aim to perpetuate its lineage through a circular cycle rather than a linear one.

3.3. Factors Affecting the Morphogenesis of Mushrooms

To inform the morphology of the oyster mushrooms (*P. ostreatus*) within a controlled environment, such as a growth chamber, it is necessary to verify the parameters that influence their growth. Understanding these parameters will help in the design and construction of the chamber used for experiments. In exploring mushroom growth, studies conducted by microbiologists and chemical engineers have been reviewed. This section, focusing on mushroom morphology, consolidates findings from various studies attempting to understand how each parameter impacts mushroom morphology.

The form of mushrooms is predominantly influenced by the specific strain of mushroom. While these criteria are generally applicable across all species, there may be variations. For oyster mushrooms, the key factors include air temperature, light, humidity, CO_2 level, gravity, substrate amount, and size (Bellettini et al. 2019). Below, the effects of these factors on the oyster mushroom morphology are examined.

Light

Oyster mushrooms exhibit phototropism; meaning they respond to light (Bayer and McIntyre 2011). In the complete absence of light, they either fail to form a cap or, at most, develop a tiny one. However, the stalks continue to grow, adopting a coral-like structure (Bellettini et al. 2019). When exposed to a blue light source, the elongation of their stalks is influenced by the light's direction. For example, when exposed to light from below, the mushrooms consistently grow downward toward the light source at all stages of development (Sakamoto 2018).

Ecovative (a mycelium technology company that designs and grows sustainable materials derived from nature) holds a patent for producing fruiting bodies of a specific shape by regulating the microclimate around the organism. They modify light intensity and frequency to in-

fluence stalk size and shapes, as seen in Figure 4 (Bayer and McIntyre 2011). The company uses a mould to restrict and direct the received light while guiding the growth of mushrooms.

Light exposure also induces colouration of the mushrooms. The pinhead browns immediately after receiving light, and reaches a brown hue that remains as it matures into a fruiting body. The colour change occurs over approximately two days in the presence of light (Sakamoto 2018).

Temperature

Temperature change is one of the triggers for the growth stages of fungi. While mycelium incubates at 22-28^{0}C, mushroom growth is initiated at 6-16^{0}C (Moore et al. 2008). These changes occur because the proteins, specifically responsible for the generation of fruiting bodies, are induced by a temperature decrease in the absence of light (Sakamoto 2018). To maintain the fructification, a temperature range between 20-25^{0}C is necessary. Lower temperatures result in a reduction of stalk height and cap size of the mushroom (Bellettini et al. 2019).

Relative Humidity

Fungi thrive in high humidity, which is also favourable for fruiting (Moore et al. 2008). Various sources achieve optimal mushroom yield by maintaining either 90-95% humidity, employing an 80-85% humidified culture room, or regularly spraying the fungi three times a day [(Bellettini et al. 2019) and (“Mycelium for Professionals” 2020)]. Stalk thickness tends to decrease with a reduction in humidity levels (Jang et al. 2003).

CO_2

A change in CO_2 concentration also triggers different stages of the fungal life cycle and affects the morphology of mushrooms. As mushrooms develop, respiration activity increases, leading to a decrease in the preferred CO_2 level. While the preferred CO_2 concentration is 2000–2500 mgL for mycelium growth, it decreases to 1500–2000 mgL for fruiting body development. If the CO_2 level remains high, the cap formation may not occur (Bellettini et al. 2019). High CO_2 concentration blocks pileus formation while boosting stalk elongation because the cell wall component, R-glucan, is affected by an elevated CO_2 level (Sakamoto 2018).

Gravity

Many mushroom stalks exhibit negative gravitropism (Moore 2010a). Gravitropism refers to the movement or growth of plants in response to gravity. When the organism grows in the opposite direction of gravity, it is called negative gravitropism (Sakamoto 2018). The cap itself may not be necessary for gravitropism. However, the bending of the stalk occurs at the upper region closest to the cap, showing negative gravitropism (Sakamoto 2018).

Substrate Amount

The amount of substrate directly influences the abundance of mushrooms blooming. Inoculating the fungi in small bags with reduced substrate results in diminished cap and mushroom sizes. In contrast, employing larger bags provides mushrooms with the opportunity to grow larger and extend further (Bellettini et al. 2019).

Substrate Size

The size of substrate (that provides the necessary nutrients for the mushrooms to grow) affects both the mushroom growth rate and the duration of growth. Straw ground sizes ranging from 0.5-2.5 cm result in a higher mushroom growth rate compared to chopped straw pieces measuring 2.5-5.0 cm (Zhang, Li, and Fadel 2002). Mushrooms exhibit faster growth cycles with ground substrate compared to those with chopped straws. It is important to note, however, that having the smallest substrates does not necessarily increase fruiting. If the particles are too small, it can lead to a decrease in mushroom yield (Zhang, Li, and Fadel 2002).

In summary, mushrooms try to adapt to the environment they inhabit in order to sustain their life through reproduction (Sakamoto 2018). Pursuing this objective, they take forms that optimise the dispersal of spores. Consequently, controlling the microclimate in which they live influences their morphology, including aspects such as cap and stalk shape, density, surface finish, and fibre orientation (Bayer and McIntyre 2011). As illustrated in Table 1, many variables have an impact on their morphology. Depending on the production purposes, these factors can be altered, akin to applying forces on the organism. Given the considerable morphological flexibility of fruiting bodies, they adeptly respond to these forces, adapting to challenging conditions (Moore 2010b). In conclusion, the abundance of influencing factors and the inherent developmental plasticity of mushrooms provide the designer with a great ground for experimentation. This thesis aims to benefit from these advantages by exploring various aforementioned factors.

Table 1

Summary of the ideal growth conditions for P. ostreatus development.

	Mycelium Formation	Pinhead	Fruiting Body Formation
Duration	0-2 weeks	3 weeks	4 weeks
Temperature	22-28^{0}C	6-16^{0}C	20-25^{0}C
Light	dark	500 lx (blue light)	500 lx (blue light)
Humidity	85-90 %	90-95 %	80-95 %
CO_2 level	2000-2500 mg L	1500–2000 mg L	1500–2000 mg L
Food source	Lignin	Cellulose, hemicellulose	Cellulose, hemicellulose
Substrate size	<0.5 cm	0.5 - 2.5 cm	0.5 - 2.5 cm

4. Methods

4.1. Preliminary Material Experiments

The aim of the preliminary tests were (i) to assess the feasibility of cultivating mushrooms in various forms by a designer, and (ii) to gain insights into the design aspects of a tool, such as a growth chamber, that enables the parametric growth of mushrooms. It is known from the literature that mushroom morphologies can be altered through environmental changes, a process that is well-explored by microbiologists and mushroom producers. However, the key question here is whether designers, lacking cultivator experience, can successfully cultivate mushrooms in diverse morphologies. In this thesis, the architect's interest in mushroom cultivation contributes to the exploration of biodesign and biofabrication strategies, incorporating digital design tools for living materials and innovative New Materialistic approaches to architecture. Despite designers not being experts in mycology, they may face unforeseen challenges during the experimental setup in a scientifically rigorous manner. To have insights into the organism, architects can design preliminary experiments using an iterative approach. These preliminary tests are valuable in understanding the organism's developmental plasticity and its potential for visible morphological changes on a macro scale due to environmental changes. Anticipating these changes in advance helps save time and resources before prematurely embarking on the main design experiments.

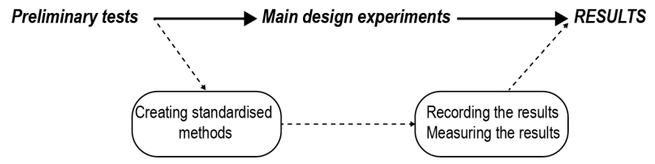
Therefore, preliminary tests are required to understand how to set up the main experiments. They are a natural part of the Research through Design (RtD) studies, allowing designers to interact with the living material. These studies create opportunities to observe and test the organism's behaviour in advance, facilitating the early identification of potential problems. This proactive approach enhances efficiency in the main experiments, particularly in new studies, by saving time and energy.

Preliminary tests also helped to define a method for recording, measuring, and analysing the results of the design experiments, as no previously used standardised method was available. Therefore, there was a need to consider and find potential strategies for measurement, enabling both quantitative and qualitative contrasts between morphologies. Three-dimensional scans were used to compare size and form, providing a visual representation of the general morphology. It helped to generate quantitative measurements, including cap size, stalk length, and curvature. However, when it came to describing differences in mushroom texture, there was not a satisfactory quantitative method. In response, macroscopic and microscopic imaging were used to present data on how changing conditions influenced these characteristics. In short, the collection, analysis, and synthesis of qualitative and quantitative data necessitated the development of new material analysis strategies, based on the insights gained from preliminary tests (Figure 5).

Three preliminary tests were carried out before initiating the main experiments. Each test contributed to a more comprehensive understanding of how to organise the main experiments to achieve reliable results in a scientific manner.

Figure 5

The diagram of the stages taken for design experiments.



- Test 1 was set up in order to see if it is possible to grow mushrooms in different forms.
- Test 2 was set up in order to understand how to design a growth chamber and how to measure the results of the experiments effectively.
- Test 3 was set up to perform final checks before running the main experiment, ensuring all necessary parameters were in place.

I. Test 1: Varied Mushroom Morphologies

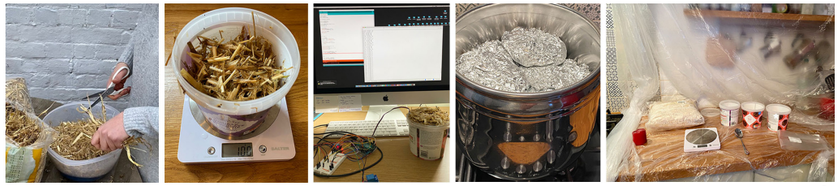
The first test aimed to understand if it is possible to achieve different mushroom morphologies. This experiment took place at home, during the beginning of the first Covid-19 lockdown restrictions in April 2020, as seen in Figure 6.

20 grams of chopped straw with a 55% moisture level were sterilised in a pressure cooker for 30 minutes. Following sterilisation, the straw was mixed with 6g of oyster mushroom spawn purchased from GroCycle-UK. The resulting mixture was then kept in 300-ml glass jars and stored in a dark space (in my closet) at room temperature for a period of three weeks.

After the initial three-week period, each jar was placed under different conditions, as seen in Figure 7. The various stages of Jars 1 through 6 are listed, as follows:

- Jar 1 was covered by a plastic bottle for 12 hours and left open for ventilation for the remaining 12 hours of the day. This was a very crude test involving modulation of ventilation.
- Jar 2 was covered by a plastic bottle on top of a humidifier. The space was kept humid, but not ventilated. The humidifier was working for an hour daily.
- Jar 3 was similar to Jar 1, but the jar remained consistently covered by a plastic bottle without any ventilation or humidification.
- Jar 4 was kept warm on a radiator with a tightly sealed lid at all times.
- Jar 5 was kept on a table rather than a radiator, with a tightly sealed lid at all times.
- Jar 6 was the same as Jar 5, but was rotated 90^0 onto its side to explore potential effects related to gravity.

The mushroom growth was monitored for ten days. It was realised that reaching different



a

b

c

d

Figure 6

Photographs of a. preparation steps, b. kitchen lab as the inoculation area, c. incubation space, and d. the growth spaces.

Figure 7

Photograph of the preliminary experimental set-up (left column) and mushroom growth on Days 21, 25, and 29.

Figure 8

Photo showing the effects of CO_2 levels on mushroom morphology (Jar 1 - left and Jar 4 - right).

Figure 9

Photography of the squashed mushroom in a glass jar.

mushroom morphologies was possible even through basic and imprecise methods. Despite the lack of scientific rigour in this particular test, it demonstrated that mushrooms exhibited distinctive characteristics under various environmental conditions. As seen in Figure 8, ventilation played a significant role in influencing cap size, stipe texture, shape, height, and quantity. Unfortunately, little else could be gleaned regarding other conditions from these preliminary tests. (The concise description of each jar is omitted as it did not yield significant results. The focus was on obtaining quick visual outcomes for subsequent in-depth exploration rather than providing exhaustive details for each jar, which were considered relatively unimportant.)

It was observed that removing substrates from the container in future experiments is crucial. Otherwise, mushrooms would continue to grow beneath the substrates inside the container, without exposure to the air, as seen in Figure 9. The jar starts to act as a mould, constraining and shaping the mushroom growth, which contradicts the objective of this study. Moreover, using a glass jar to keep the substrates proved unsuitable, as the glass's rigidity made it challenging to extract all of the substrates in one piece. Therefore, malleable plastic containers were used in subsequent tests, facilitating the easy removal of mushrooms. These containers were removed after three weeks to allow unrestricted mushroom growth from all sides, without constraints imposed by a mould.

II. Test 2: Substrate Allocation in the Growth Chamber

To achieve reproducible results for the design experiments influencing the mushroom morphologies through environmental changes, a digitally controlled growth chamber was aimed to be used as a tool. The objective of Test 2 was to understand how to design a chamber, decide the suitable substrate allocation for mushroom growth within the chamber, and assess whether any adjustments, such as changes in size, geometry, or distance between samples, were necessary before conducting experiments. As part of this assessment, humidity levels were utilised in a preliminary test run.

Triplicates were prepared for testing the effects of three different humidity levels, resulting in a total of nine samples. A blend of 80g chopped straw, 40g wood shavings, and 5g coffee grounds was sterilised in a pressure cooker for 30 minutes. Subsequently, water was added until the mixture reached a 55% moisture level. The moisture was measured through the DF Robot V1.0 soil moisture sensor. The prepared mixture was then inoculated with 45g of oyster mushroom spawn from GroCycle-UK, all under consistent conditions (sealed plastic pint cups, in the dark, at ambient temperature). After a three-week incubation period, each set of substrate triplicates, now bound together by mycelium, was extracted from the plastic cups, and transferred to a plastic box, serving as a controlled environment.

The humidity, selected as the variable for this controlled experiment, was monitored using a DHT11 air humidity sensor and regulated through a humidifier. Using a code that programs the Arduino, ambient conditions were set to 75%, 85%, and 95% air humidity for the three separate chambers, respectively. The remaining variables were kept constant: light expo-

sure for 4 hours per day and temperature at 22^{0}C.

A difference in the curvature of the mushroom stipes was observed corresponding to different levels of humidity, as seen in Figures 10 and 11. However, the results were not reliable due to the problems observed during the growth process. The first problem concerned the homogeneity of the substrate mixtures. The straw length varied at different points in the mixture, resulting in an uneven overall inoculation of the samples. There was an uneven mycelium coverage. Accordingly, less mushroom growth occurred in areas with larger substrate sizes, highlighted in circles 1 and 2, in Figure 12. As mentioned before, substrate size is one of the variables that affects the mushroom size. Thus, the resulting uneven mushroom growth corresponding to varying substrate sizes indicated that, while Test 2 was conducted, all variables except humidity were not maintained consistently. To solve this problem, future tests will involve a more uniform cutting of straw using a blender.

The second problem in this setting was the excessive amount of substrates. The sample sizes were too large to accommodate triplicates in the growth chamber, leading to insufficient space for mushrooms to grow without coming into contact with each other or the chamber walls. Consequently, the overcrowding of the chambers may had led to the formation of microclimates, although this has not been conclusively proven. There was not any mushroom growth on the sample sides facing each other, as seen in Figure 13, which could be as a reasult of oxygen shortage in between the samples.

The third problem was the geometry of the samples. It was difficult to analyse the mushroom morphology as the mushrooms grew on both curved and flat surfaces. To solve the problem of size and geometry, a decision was made to use smaller flat packs in future tests. This choice aimed to minimise variance across different points of each sample.

The test was replicated with feedback from the previous trial, this time using 10x10x4-cm plastic containers as moulds. In comparison to the previous geometric setup, a more obvious curvature difference between the mushroom stalks and cap inflexion was observed, as seen in Figures 14 and 15. The sample size provided was suitable for the growth chamber, and the flat box geometry helped in collecting comprehensive results, including mushroom

75% *85%* *95%*

Figure 10
Close-up photography depicting the curvature of mushroom stalks at various humidity levels during Test 2, on Day 27.

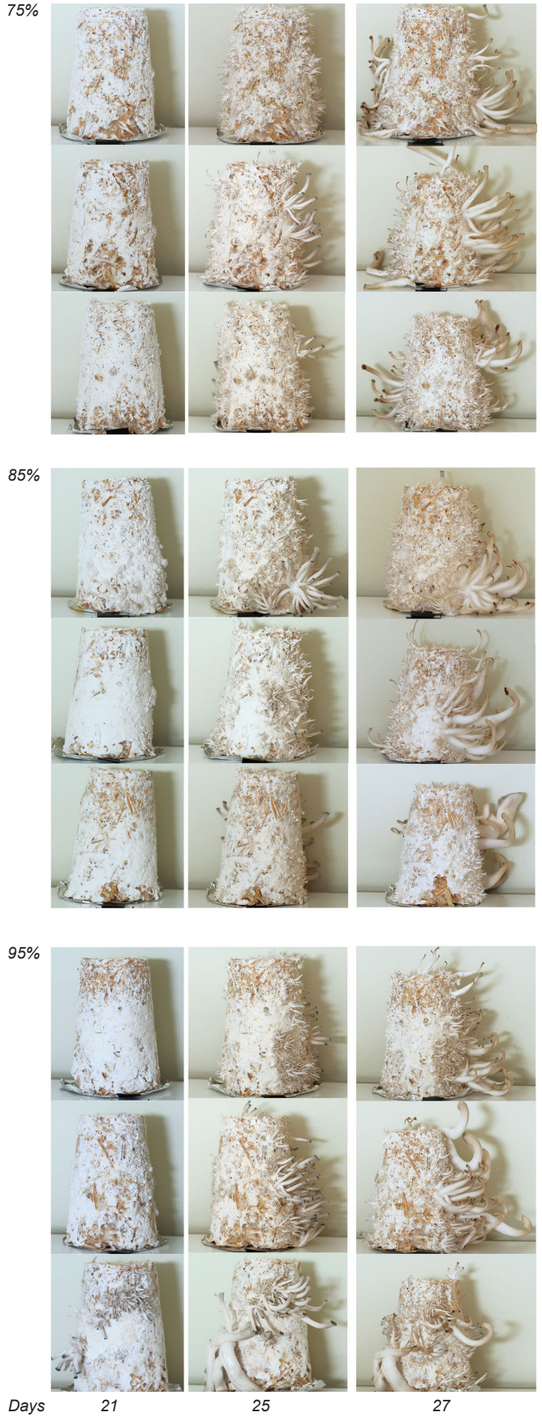


Figure 11
Photography depicting the morphology of mushrooms at various humidity levels during Test 2, on Days 21,25 and 27.

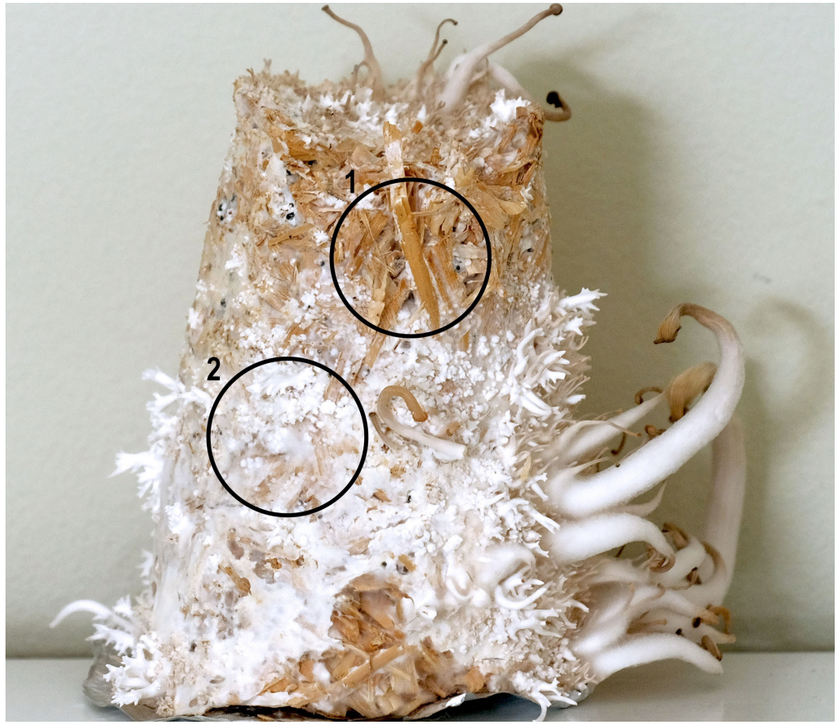


Figure 12

Uneven mycelium growth in circles 1 and 2.

Figure 13

Mushroom growth observed exclusively on the sides that are not in direct contact with another sample.

location, size, length, texture, and the number of sprouts, by providing a clear reference plane for measurement.

Even though many problems were fixed in this trial, there was still an overlooked detail. The mycelium was more evenly distributed, but still had not completely covered the sample. A longer inoculation period was required to achieve full coverage of the substrates. However, due to time constraints, the mycelium-substrate composites were taken out of the mould after the minimum time required for fruiting body formation.

III. Test 3: Setting Up The Duration Of Experiments

With this last preliminary test, the duration of the experiments was considered. As mentioned in the growth process (Table 1), mycelium starts forming pinheads after three weeks, and these pinheads transform into mushrooms a week later. On Day 27, mushrooms reach their full size, as seen in Figure 16, and after that, they begin to wilt (often seen after Day 30). Therefore, the morphology studies were carried out based on the condition of the mushrooms on the 27th day.

Another realisation from this test was that the horizontal positioning of the samples posed a problem. The samples were sitting on their widest surface, which caused a moist area. Consequently, pinheads started to grow and form mushrooms on these surfaces, as seen in Figure 17. This indicated that the bottom of the growth chamber was starting to act as a mould. Although preventing the specimens from touching any surface was not feasible, keeping them upright on the smallest plan in future experiments would help minimise physical contact.

Figure 14

Close-up photograph showcasing the observation of mushroom curvature in flat-pack moulds under different humidity levels.

75% *85%* *95%*

Figure 15

Mushroom curvature variations on days 21, 23 and 27 in flat-pack moulds correlated with humidity levels.

Figure 16

Photographs of five samples taken on Days 23, 27, and 29, illustrating the point at which mushrooms reach their full size and commence wilting.

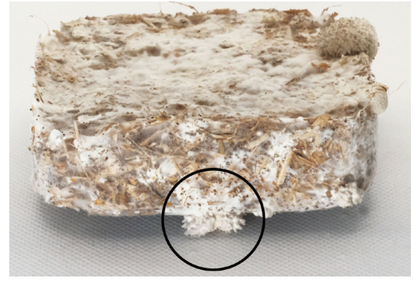


Figure 17

Close-up photograph showing mushrooms growing between points where the sample chamber makes.

IV. Inferences

Preliminary experiments have guided the decision to pursue mushrooms rather than mycelium. These initial investigations helped to see dominant parameters that significantly influence mushroom morphology. Moreover, they bolstered my confidence and familiarity with working with mushrooms. Each preliminary experiment contributed to a more profound understanding of how to set and progress to the main experiments. They helped in determining and implementing the following:

- The choice of container material for keeping substrates.
- The importance of achieving aggregate homogeneity.
- The optimal geometry for incubating the substrate within the container.
- The duration of the experiment.
- The positioning of the samples in the growth chamber.

The main experiments and the digitally controlled growth chamber were designed in light of these findings from the preliminary tests. Square plastic containers, 10x10x4 cm in size, were used to keep substrates in, and they were easily removed after the initial three weeks. Straw was homogenised by blending in a Nutri Ninja Blender and Smoothie Maker 900W for a period of five seconds. Wood shavings and coffee grounds were not blended since they were standard in size. Following the extraction of samples from the plastic containers, they were vertically positioned within the growth chamber. The experiments were concluded on the 27th day.

4.2. Designing a Digitally Controlled Growth Chamber

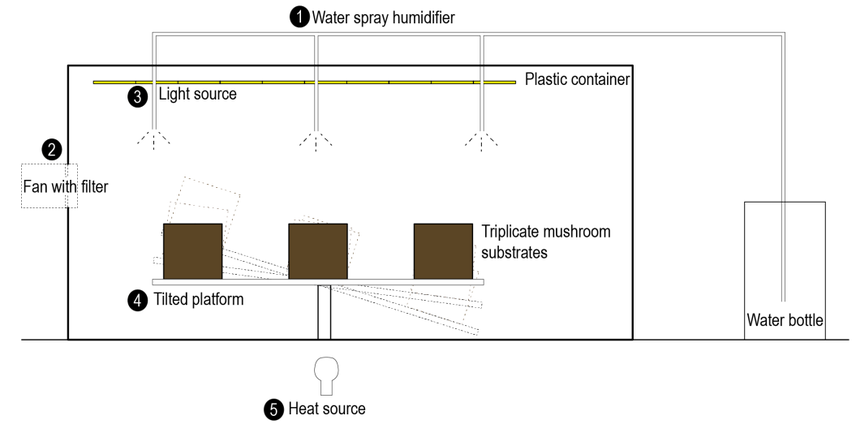
After the preliminary tests were completed, the next phase involved conducting more intricate scientific experiments to explore the profound relationships between environmental parameters and morphologies. To do that there is a need to build an environment chamber for precise control. The preliminary tests have proven valuable in confirming the effectiveness of a plastic box regulated by sensors and actuators. These initial experiments not only validated the functionality, but provided insights for refining the chamber's design, the sample sizes, and geometric parameters established in accordance with the predetermined chamber design. Furthermore, the use of digital tools might help to facilitate the autonomy of biologically active organisms instead of exerting dominance over them. This gives rise to new symbiotic relationships. Blending digital and biological systems helps to address new questions in interactive design. This dynamic interaction forms and nurtures organisms, converting them into a programmable material system, and functionalises them for specific applications (Zolotovsky 2017).

Based on Mario Carpo's categorisation of making types, the use of a digitally controlled chamber falls within the realm of digital making. Carpo's classification of making (hand, machine, and digital) provides a valuable framework for understanding diverse approaches to fabrication tools. Hand making includes arts and crafts, where variations exist. Machine making represents an industrial approach that yields numerous identical copies of the final product. Lastly, digital/computational making integrates tools from machine making, such as 3D printers, CNC machines, and robotic tools, while enabling customisation. It combines the advantages of both hand and machine making, allowing fine reproducibility. Digital making is an interactive and collaborative process, as it allows the manipulation of input parameters that transform numeric variables into objects (Carpo 2017). Adapting digital tools for fabricating living materials for design purposes does not entirely eliminate human authorship. Instead, it enhances the biofabrication process, making it more precise and interactive.

Living materials, which respond to environmental changes, present unique challenges compared to obedient materials . When fabricating them using digital tools, it is essential to adjust and optimise these tools to meet the specific requirements of the material at hand. Therefore, before designing a digitally controlled growth chamber, parameters affecting mushroom morphology were carefully considered. The variables impacting mushroom morphology include light, temperature, humidity, CO_2 levels, gravity, substrate amount, and substrate size. Although not all of these variables can be controlled by a digital system, environmental factors can be monitored and modified. Therefore, initially, the following five parameters were selected as modifiable variables: light, temperature, humidity, gravity, and CO_2 levels.

Figure 18 illustrates the initial sketch of the growth chamber. In practice, the substrate triplicates were placed in a closed container, and the container was regulated through an Arduino, along with various sensors and devices. However, during the system construction, the decision was made not to connect the light source (Figure 18-3) and the tilted platform (Figure 18-4) to the Arduino. While they could be integrated into the system, there was no necessity for sensors concerning these variables. As the light can simply be on or off in this system, independent of other variables, and does not require a sensor, it was more efficiently controlled using a timer switch. Furthermore, the platform did not need feedback, was manually controlled and positioned, as constructing a mechanically feedback-less system was more practical. The parameters requiring feedback, such as temperature, humidity, and CO_2 levels, were therefore modulated using Arduino sensors and actuators.

Figure 18
The Initial sketch of the growth chamber.



Ultimately, the DHT11 air humidity and temperature sensor, SEN0219 infrared CO_2 sensor, V1.0 soil moisture sensor, and HC-SP04 ultrasonic distance sensor were used, as shown in Figure 19. These sensors were connected via an Arduino UNO to a computer, together with the devices, including a 12V DC fan (to regulate CO_2 levels), ASAKUKI humidifier with 500ml water tank (to regulate humidity), and 75-watt heat bulb (to regulate temperature). The sensors and devices were all integrated into a clear plastic container (48x36x30-cm in size), as shown in Figure 20. As mentioned before, the blue light source and the positioning of the samples were controlled by the designer rather than Arduino. A 450 nm LED blue light source was turned on/off by the designer at specific times of the day. To prevent the chamber from exposure to daylight, a cardboard box was placed over the plastic container. For the positioning, small plastic kitchen boxes were used to arrange the samples at different angles. Three replicates of the chamber were built, as seen in Figure 21, to help run the experiments simultaneously, thereby making more efficient use of time.

Apart from the directional change in gravitational force and substrate amount, all other environmental parameters were digitally controlled. It was more time and cost-efficient to eliminate the digitally tilted platform concept, given the constraints of the PhD. The resulting chamber could sense using Arduino sensors and respond accordingly through actuators based on the defined algorithm.

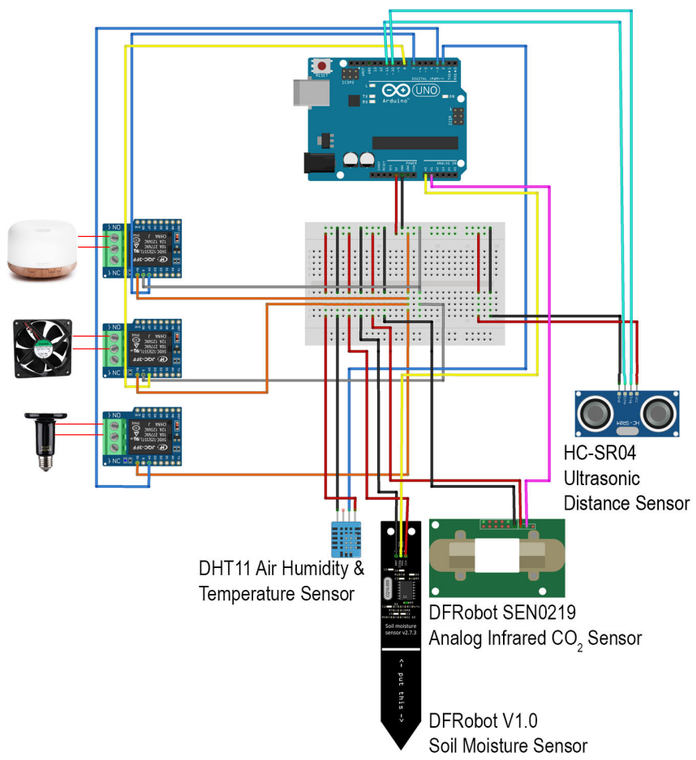


Figure 19

Diagram of the wire connection of Arduino, sensors, relay modules, and actuators that were used for the growth chamber.

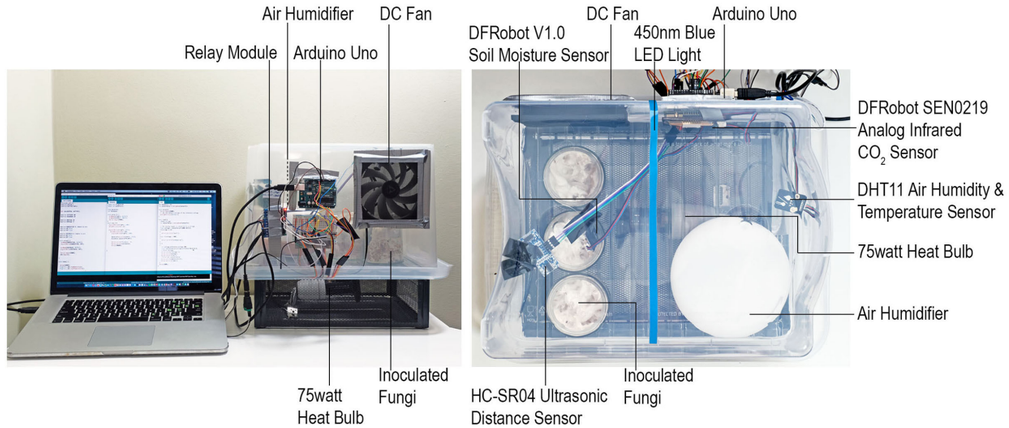


Figure 20

The digitally controlled growth chamber and its components.

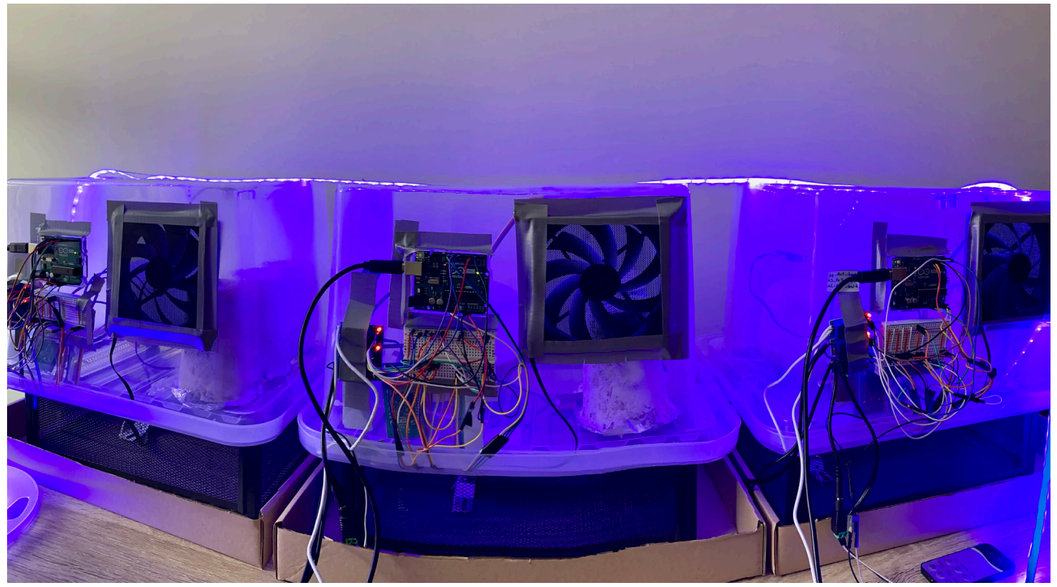


Figure 21

Photography of the chambers in triplicate, this is done to denote that a laboratory experiment was done three times to ensure accuracy and precision of the results.

The system used in the growth chamber, as seen in Figure 22, principally operates using 'on' and 'off' codes. The actuators, which are the command-implementing devices, activate or deactivate based on sensor readings and input values. The input values are the environmental conditions desired by the designer. Actuators turn off when the specified conditions are met, and this information is detected by the sensors. For example, the humidifier continues to operate until it reaches the designated humidity level. In essence, all of the devices within the growth chamber function similarly to a thermostat, ensuring precise control and maintenance of the specified environmental parameters. As a result, fruiting bodies grow and form in response to the environmental conditions.

All the sensors, except for the distance sensor, are designed to measure the environmental conditions within the chamber. The primary responsibility of the distance sensor is to measure the growth of mushrooms positioned directly beneath it. Initially, the intention was for the distance sensor to provide feedback for environmental controls, allowing for the regulation of conditions that either inhibit or promote growth. The envisioned scenario involved, for instance, the distance sensor triggering the actuators to deactivate when mushrooms reached a certain height. This approach aimed to create a smart, and cybernetic system, wherein a sensor monitored mushroom growth, influencing the actuators. However, this integration could not be achieved for two reasons.

Firstly, the mushrooms did not consistently grow directly beneath the sensor, even after manual repositioning; their growth was often irregular. Therefore, a sensor with a camera module seemed more suitable for accurately tracking changes in mushroom growth. Secondly, the actuators only responded to commands from environmental input (through CO_2 and temperature humidity sensors). To receive input from the distance sensors, adjustments to the code were necessary during the growth process. The distance sensor encountered several issues and, ultimately, was not utilised in the experiments.

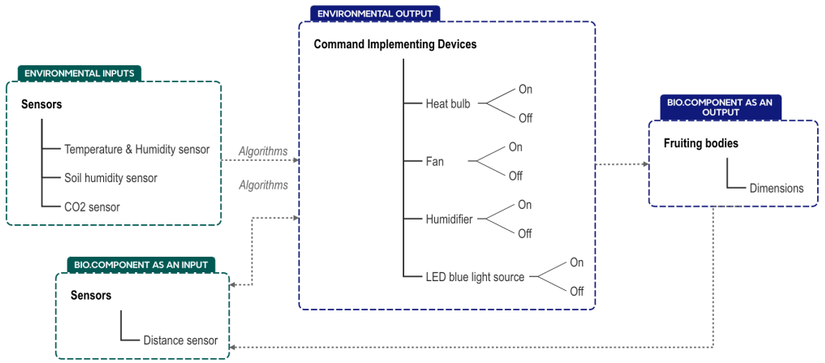


Figure 22

Diagram of relations for the growth chamber.

4.3. The Main Design Experiments

To guide the growth of mushrooms within a digitally controlled growth chamber, an exploration into the impact of humidity, CO_2 levels, gravity, and substrate amount on mushroom morphology was investigated using a scientific method. Although gravity and substrate amount are not variables controlled by the chamber, they influence the mushroom size and curvature. More dramatic results were expected by changing the substrate amount and gravity. Therefore, they were chosen as variables, which would be manipulated by the chamber.

A scientific method was used in experiments as mentioned in the methodology chapter. A direct comparison of the specific conditions helped in understanding the influence of each condition on the organisms' morphology. To achieve this, only one variable was changed at a time, while all other variables remained constant. The ambient conditions required for fruiting body maturation and mass cultivation for agricultural purposes are summarised in Table 1 (Jang et al. 2003) (Watkinson, Boddy, and Money 2016). These results were utilised to give some scope in making decisions while setting up the experiments.

All experiments were conducted in triplicate to help detect the 'typical' mushroom morphology formed each time under a single condition. Each set started with the same substrate ratios (25% of strawbale, 25% of wood shavings from Pets at Home, and 25% of coffee grounds), which were sterilised in an autoclave at 121^0C for 15 minutes. The substrates were prepared at home and taken to the lab for sterilisation. This mixture was then seeded with 25% of oyster mushroom spawn from GroCycle-UK in the lab, and then taken home where they were placed under the same conditions: in sealed plastic boxes, in the dark, at ambient temperature (in a drawer). After three weeks, they were exposed to different environmental conditions for eight days, controlled by altering one parameter at a time (Table 2). The process of bringing the samples back and forth was necessary due to the COVID-19 pandemic. Many of the steps could be undertaken at home, but the sterile environment and autoclave of the lab were too challenging to consistently recreate at home.

Table 2

The summary of the variables used for four experiments.

Variables	**Experiments**			
	[1] Humidity	**[2] CO_2**	**[3] Gravity**	**[4] Sub. Amount**
Humidity (%)	95, 85, 80, 75	80	75	80
CO_2 (ppm)	2000	1000, 3000, 5000	5000	3000
Substrate (g)	55	55	55	40, 80,120,160
Gravity (deg.)	90^0	90^0	90^0, 135^0, 180^0	90^0
Light (nm)	4h, 450 nm	4h, 450 nm	4h, 450 nm	4h, 450 nm
Temp. (0^c)	20-22^{0}C	20-22^{0}C	20-22^{0}C	20-22^{0}C

I. The humidity experiments

The variable humidity conditions in this controlled experiment were measured using a DHT11 air humidity sensor, and adjusted with a humidifier. In this way, the ambient conditions could be adjusted to 75%, 85% and 95% air humidity by utilising code which programs the Arduino. The other variables, which were kept constant, were CO_2 level at 3000 ppm, light exposure for 4 hours per day at the same time of the day and temperature at 22^0C in this set of experiments.

II. The CO_2 experiments

In this set of experiments, the CO_2 levels were tested at 1000 ppm, 3000 ppm, and 5000 ppm. These were achieved with the help of the fan and SEN2019 Analog Infrared CO_2 sensor in the growth chamber. The other variables, which were kept constant, were air humidity at 80%, light exposure for four hours per day at the same time of the day, and temperature at 22^0C in the experiment.

III. The gravity experiments

In this experiment, the angle of growth was tested. The effect of gravity upon the growing mushroom was adjusted as a means of support by using the aforementioned plastic containers. After being removed from the containers, the mycelium tiles were kept at 90^0, 135^0 and 180^0 angles, as seen in Figure 23. The samples with 180^0 angles were positioned on a box. Lifting them prevented moistening and mushroom growth on contact surfaces. The experiment was repeated under a 2000 ppm CO_2 level. As such, it was possible to see the effect of gravity on caps in different sizes.

IV. The substrate amount experiment

In this set of experiments, the effect of substrate amount on mushroom size was tested. Mycelium in the amounts of 40g, 80g, 120g, and 160g, along with various substrates, were mixed in the ratio of 25% of strawbale, 25% of wood shavings, 25% of coffee grounds, and 25% of mushroom spawn, as mentioned before. All mixtures were kept in a 10x10x3cm plastic box and covered with aluminium foil with a 4x4 cm hole in the middle of one of the widest surfaces, as seen in Figure 24. The aim of guiding the mushroom growth from a single opening was to limit the number of fruiting bodies, thus preventing overcrowding, to retain focus on the size of the mushrooms.

Figure 23

The positioning of the mushrooms in the gravity experiment .

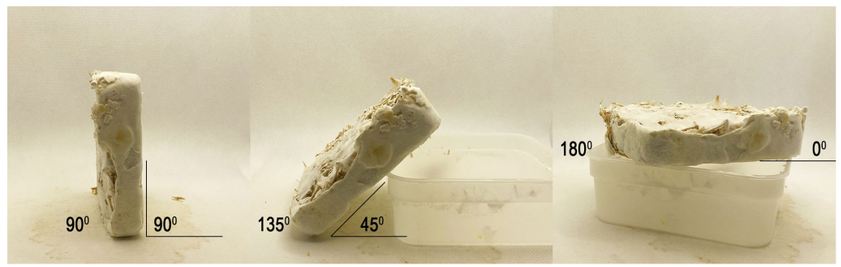


Figure 24

Photograph showing the preparation of samples for the substrate amount experiment.

4.4. Recording the Results

The mushroom morphology was documented on Day 27 through various techniques: photography (Fujifilm X-T2 with 80mm lens), microscopy (Dino-Lite digital microscope at 70X magnification), and 3D scanning (EinScan-SE desktop scanner) (Figure 25). Photography helped to analyse the overall form and general tendency of mushrooms. Microscopic images, on the other hand, captured intricate details, smaller features, and non-measurable characteristics such as surface texture and colour. The images from photography and microscopy generated the qualitative data for this study and helped to compare mushroom formations in both micro and macro scales.

The 3D scanning process allowed the translation of the mushroom forms into digital models using Rhinoceros (Figure 26). This approach helped to obtain more precise measurements of the angles and dimensions of the caps and stalks without causing damage to the organisms. It also became possible to overlay the triplicate three-dimensional scans of the mushrooms grown under identical environmental conditions. This overlaying method allowed for a comprehensive comparison and contrast of morphologies through the digital files. Ultimately, it helped in visually representing the average morphology for each condition.

4.5. Measuring the Results

Three-dimensional scans helped to analyse the overall mushroom forms by allowing for the digital measurement of the dimensions of the caps and stalks. To measure, the largest mushroom from each replicate was selected as the most mature specimen as a sampling method. If a substrate block produced multiple mushrooms, only the largest one was considered, while smaller mushrooms were disregarded. In total, three mushrooms for each condition were used, and their mean values were calculated. Due to the difficulty of manually measuring delicate mushrooms, measurements were made digitally using Rhinoceros. Other than for measurement purposes, three-dimensional scans were used for visual comparison by overlaying all the samples. While doing so, instead of only using the largest mushroom on each sample, all mushrooms that were sufficiently large to be scanned were included. The location of the measurement points for each specimen was standardised, as follows (Figure 27):

- Cap size and stalk length were measured using curved lines. To measure the cap size |AB|, point A was selected arbitrarily on the cap edge, and point B was located on the opposite side of the edge/point A. To measure the stalk length |EF|, Point-F was selected as the bottom of the stalk and Point-E was selected as the lowest mid-point of the cap.
- The angle of the cap curvature (D^0) was measured by:
    1. Drawing a line between the lowest and highest point on the cap edge.
    2. Measuring the angle between this line and the x-axis (parallel to the ground).
- The stalk curvature angle (G^0) was measured by drawing two lines parallel to the stalk (one from underneath the cap, the other from the base of the stalk) and measuring the angle between these two lines.

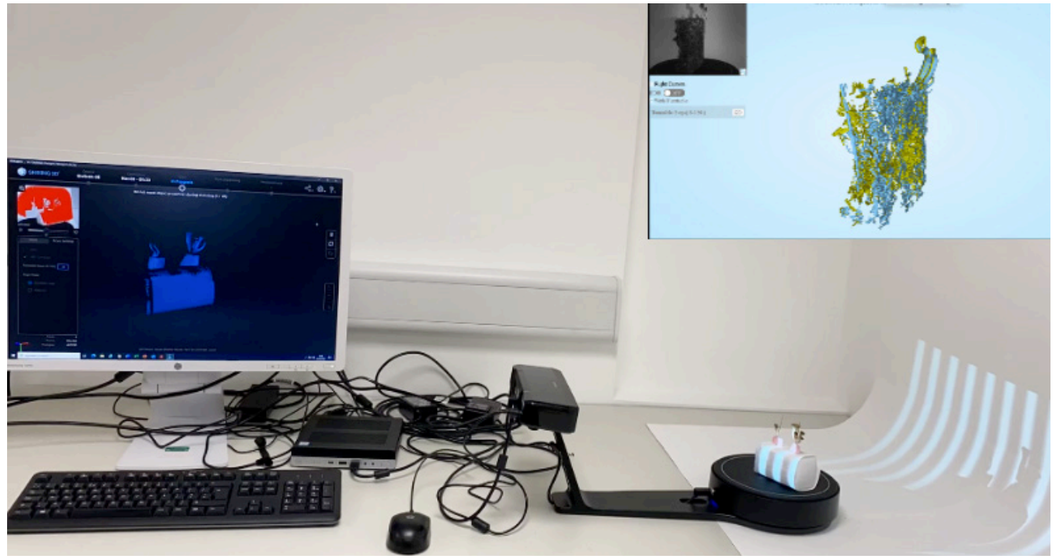


Figure 25

A photograph capturing the 3D scanning process of mushrooms using the EinScan-SE desktop scanner, with an additional corner image displaying a screenshot from the software, depicting how the scan appears on the scanner's interface.

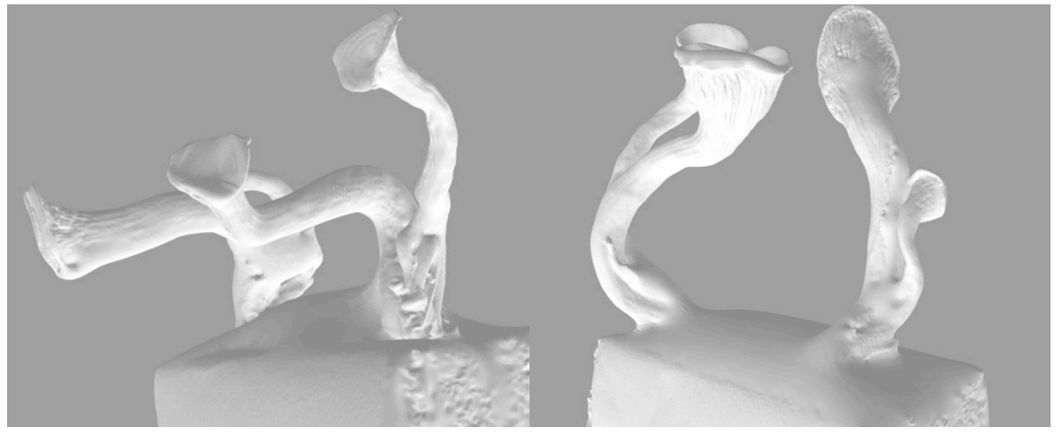


Figure 26

A screenshot from Rhinoceros showing the 3D mushroom scans in .stl format.

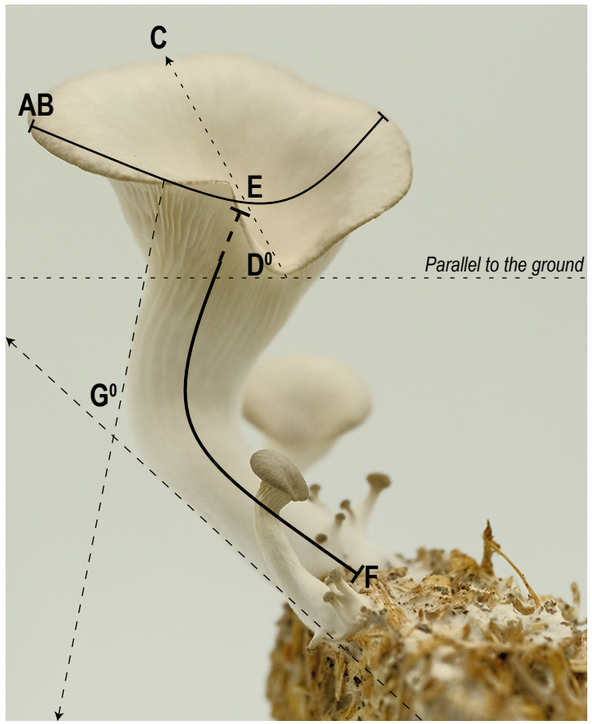


Figure 27

A marked photograph showing the locations of the measurement points.

A *A point on the cap edge*
B *The opposite of point A on the cap edge*
|AB| *Cap size*
C *The line between the lowest and highest point of the cap*
D⁰ *Cap curvature angle*
E *Lowest middle point of the cap*
F *Bottom of the stalk*
|EF| *Stalk length*
G⁰ *Stalk bend angle*

4.6. Calculating the Standard Error

To ensure scientific rigour, the research was run with three replicates to validate the reproducibility of the results with each controlled variable. The use of triplicates helped compare and confirm the observed results, facilitating the identification of the 'typical' fungal morphology formed under a single condition. This involved calculating the mean average of fungal sizes, volumes, and angles that contributed to the overall form.

During the experiments, these outcomes were quantified, and the standard deviation (SD) between them was calculated. Measuring the amount of variability helped in determining the standard error (SE), representing the difference between a sample and the average. "**The standard error of the sample mean** depends on both the standard deviation and the sample size, by the simple relation SE = SD/√ (sample size). The standard error falls as the sample size increases, as the extent of chance variation is reduced—this idea underlies the sample size calculation for a controlled trial, for example" (Altman and Bland 2005). The standard error served as an indicator of certainty; therefore, it was important for precision as a criterion in evaluating the reliability of the proposed fabrication method.

In any experiment, it is ideal to achieve low deviations between samples and to have consistent results. However, outliers may occasionally arise, causing a sample to deviate from the expected standard. In this research, when an outlier was detected, the experiment was repeated to ensure consistent results, and the outlier was subsequently excluded from the analysis phase. If, during the repeated experiment, the organism performed similarly to its previous behaviour, it was not deemed an outlier, and was included in the final results. While discarding results was the preferred approach, it was accepted as necessary in this research where outliers did not follow the expected pattern, thereby skewing the overall results.

In experiments involving mushrooms, the organisms exhibited inherent autonomy, leading to diverse sizes. From the perspective of a designer, acknowledging this, it was accepted that smaller sizes would be discarded and excluded from the final analysis. This decision to include only the biggest mushroom was based on the understanding that such forms would not provide meaningful or relevant information to the objectives of the research.

5. Results

I. Results of the humidity experiment

The results evaluate the performance of the growth chamber, examine mushroom morphologies under various environmental conditions, and explore the bio-digital fabrication method as a concept approach.

Humidity influenced the curvature of cap edges and the stalks, as seen in Figure 28 and Table 3. The replicates grown in the in-between conditions exhibited in-between morphologies. There was a tendency for certain variables to increase in response to higher humidity levels. For instance, stalk and cap curvature tended to increase with rising humidity. On the other hand, some variables exhibited no correlation, such as stalk length, with changes in humidity.

However, the cap sizes seem smaller in 80% humidity, than the mushrooms grown in 75%. That could be because the two outlier mushrooms grew bigger than expected and raised the average value, although there is not enough data to prove that. The texture of stalks and the depth of gills are qualitative results. It can be observed from Figure 28 that, in 80%, gills were shallower, and stipes were hairier than in 75%.

Table 3

Measurements from the humidity experiment. The highlighted text indicates a tendency (by an increase or decrease) in the change of variables.

		95%			85%			80%			75%		
		Rep-1A	**Rep-2A**	**Rep-3A**	**Rep-1B**	**Rep-2B**	**Rep-3B**	**Rep-1C**	**Rep-2C**	**Rep-3C**	**Rep-1D**	**Rep-2D**	**Rep-3D**
Cap	Size (cm)	2.8	3.1	3.1	1.8	3.2	3.4	2.1	2.3	2.4	3.9	1.6	3.6
	Average size		3±0.1			2.8±0.6			2.3±0.1			3±0.9	
	Curvature	120	127	145	53	64	60	30	38	31	31	32	36
	Average curv.		130.7±7.4			59±3.2			33±3.3			33±1.5	
Stalk	Length (cm)	5.6	5	4	4	4.9	4.4	3.5	4.1	4.7	4.7	4	3.2
	Average length		4.9±0.5			4.4±0.3			4.1±0.3			4±0.4	
	Curvature	104	133	140	85	83	90	51	78	81	54	61	76
	Average curv.		125.7±2.9			86±2.9			70±7.8			63.7±6.1	
Average sprout number			2			4.3			4			3.3	

Figure 28

Photos captured on Day 27 showcase the morphologies of mushrooms under four distinct humidity levels. The images include front, side (right and middle columns), and zoomed-in views (left column).

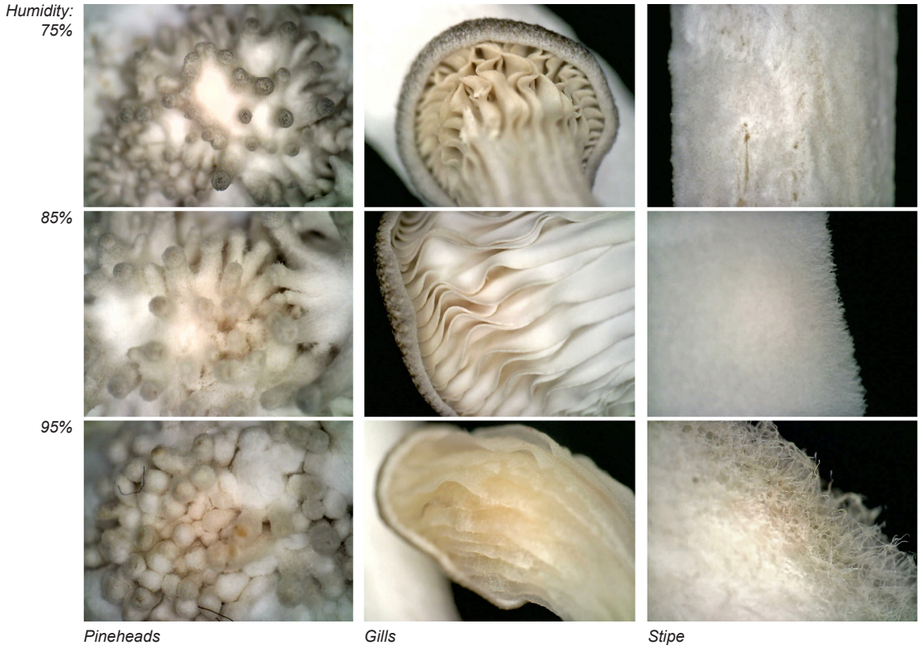


Figure 29.1

Microscopic images of different parts of mushrooms in the humidity experiment. The comparison of gills and stipes under different humility levels. Images are captured using a Dino-Lite digital microscope at 70X magnification.

Figure 29.2

Microscopic images of different parts of mushrooms in the CO_2 experiment.

II. Results of the CO_2 experiment

CO_2 mainly affected the size of the caps, as seen in Table 4. The caps grew bigger with a decrease in CO_2 levels. The stalks gtrew longer with higher CO_2 levels (Figures 29-2 and 30). However, after a certain point (approximately over 3500 ppm), their length did not increase anymore, and they became leaner. In short, it can be stated that while humidity had more influence on the curvature of the stalk, CO_2 significantly affected the cap size.

Table 4

The measurements of the CO_2 experiment. The highlighted text indicates a tendency (by an increase or decrease) in the change of variables.

		5000 ppm			3000 ppm			1000 ppm		
		Rep-1A	**Rep-2A**	**Rep-3A**	**Rep-1B**	**Rep-2B**	**Rep-3B**	**Rep-1C**	**Rep-2C**	**Rep-3C**
Cap	Size (cm)	0.4	0.2	0.1	1.3	0.8	1	2.4	2.3	2.5
	Average size		0.2±0.1			1±0.2			2.4±0.1	
	Curvature (^{0})	31	2	5	41	53	40	8	7	9
	Average curv.		12.7±11.8			44.7±5.3			8±0.8	
Stalk	Length (cm)	3.8	0.2	3	4.5	3.8	4.4	2	1.8	2.1
	Average length		2.3±1.5			4.2±0.3			2±0.1	
	Curvature (^{0})	66	79	73	44	41	39	53	61	55
	Average curv.		72.7±5.3			41±1.2			56.3±3.3	
Average sprout number			8.3			6			6.6	

Figure 30

Photos captured on Day 27 showcase the morphologies of mushrooms under three distinct CO_2 levels. The images include front, side (right and middle columns), and zoomed-in views (left column).

III. Results of the gravity experiment

As seen in Table 5, there was a tendency for the fruiting body to grow vertically, so the mushroom caps tended towards being parallel with the horizontal plane. This led to the stalks being bent from underneath the cap as they grew away from the tilted plane of the tile towards a vertical direction. As seen in Figures 31.1 and 31.2, the mushrooms grown both at 5000 (high) and 2000 (low) ppm of CO_2 presented the same behaviour, in terms of orientation. In summary, gravity affected the orientation of the caps, which led the stalks to curve accordingly, but there was no significant impact on the size of the mushrooms.

Table 5

The measurements of the gravity experiment in high CO_2.

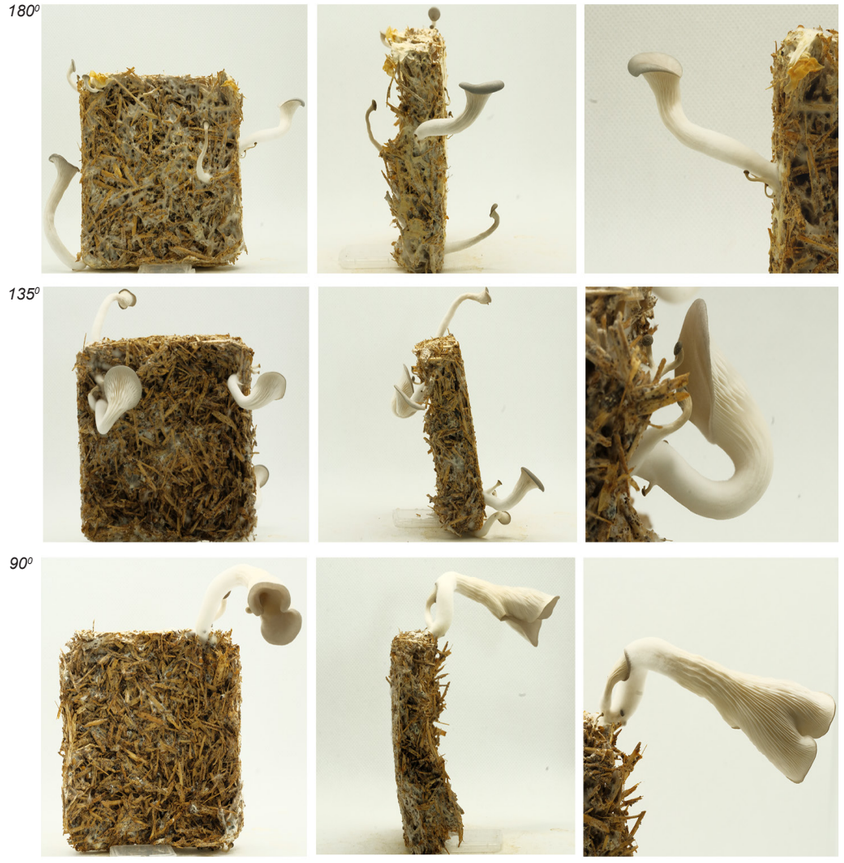
		90^0			135^0			180^0		
		Rep-1A	**Rep-2A**	**Rep-3A**	**Rep-1B**	**Rep-2B**	**Rep-3B**	**Rep-1C**	**Rep-2C**	**Rep-3C**
Cap	Curvature (^{0})	6	12	0	61	35	16	44	19	35
	Average curv.	6±2.4			37.3±10.6			32.7±10.2		
Stalk	Curvature (^{0})	44	51	57	35	23	46	23	36	0
	Average curv.	50.7±3.7			34.7±6.6			19.7±10.5		

Figure 31.1

Photos captured on Day 27 showcase the morphologies of mushrooms at high CO_2 levels (5000 ppm) and different levels of orientation parallel to the ground. The images include front, side (right and middle columns), and zoomed-in views (left column).

Figure 31.2

The same experiment was repeated under low CO_2 levels (2000 ppm).



IV. Results of the substrate amount experiment

When the mushroom sizes and the number of sprouts are compared, as seen in Figure 32, an increase in cap size, stalk length, and sprout number can be observed as the substrate amount increases (Table 6). The reason for mushroom stalks growing with different curvatures is that they curled as they came out of the hole in the foil wrap.

The variable that had the most effect on the overall size of the mushrooms was the amount of substrates. Although an increase in humidity enlarged them to some extent, the substrate amount was the main determinant. Without sufficient substrate, the mushrooms could not reach their maturity.

The samples grown in in-between conditions exhibited in-between morphologies. As we know from the previous substrate amount experiment, the size of cap edges and the stalks increased as the substrate amount increased. All the curvature measurements in the 120g mixture are somewhere between 80g and 160g substrate amounts. Although the sprout number is similar to the 80g sample, it is less than the 160g sample.

Table 6

The measurements of substrate amount experiment. The highlighted text indicates a tendency (by an increase or decrease) in the change of variables.

		40g			80g			120g			160g		
		Rep-1A	**Rep-2A**	**Rep-3A**	**Rep-1B**	**Rep-2B**	**Rep-3B**	**Rep-1C**	**Rep-2C**	**Rep-3C**	**Rep-1D**	**Rep-2D**	**Rep-3D**
Cap	Size (cm)	1.3	1.2	2.9	2.2	2.3	1.8	3.4	3.7	1.5	3.9	2.5	2.8
	Average size		1.8±2.1			2.1±0.6			2.9±2.7			3.1±0.4	
Stalk	Length (cm)	5.3	2.9	5.1	5.8	6	5.2	5.8	6.7	5.2	6.7	5.4	6.3
	Average length		4.4±1			5.7±0.1			5.9±0.4			6.1±0.5	
Average sprout number			4			6			5			9	

Figure 32

Photos captured on Day 27 showcase the morphologies of mushrooms growing in different substrate amounts. The images include front, side (right and middle columns), and zoomed-in views (left column).

V. The performance of the growth chamber

The digitally controlled growth chamber eases biofabrication for designers by correlating morphological outputs with digital data in the code. In other words, it creates a method for handling the complexity of the mushroom morphologies. However, while conducting the experiments, it was noticed that there are factors which complicate the system of the growth chamber.

The objective of using a growth chamber was to establish a stable environment within a closed system, enabling precise control over various parameters. However, this goal became more complex with the introduction of feedback relationships. The cellular respiration of the mushrooms produces CO_2, heat and water, thereby affecting humidity, temperature, and CO_2 levels in the chamber. As a result, the mushroom's ability to actively modify its surroundings requires the chamber to adapt to these fluctuations, maintaining a consistent equilibrium. Therefore, it engages in a dynamic relationship with the organism, responding to its changes in real time. This very dynamic relationship makes the chamber system a cybernetic system, as mushrooms are in constant dialogue with the digital system. Despite being an unintended outcome of the chamber's design, this dynamic relationship exemplifies its cybernetic nature.

Another complicating factor in the system is the operation of the devices; while aiming to affect one variable, it affects the entire system. For instance, running the fan reduces the CO_2 as intended, but also inadvertently affects (drops) the humidity level and temperature in the chamber. Additionally, independent from the devices, the innate relationship between environmental factors can result in unexpected changes in variable levels, which is a feature of closed systems. For instance, humidity rises when the temperature drops. Despite this interplay between the different inputs of the system, which is quite complex, all the devices work simultaneously to keep the conditions constant.

Other than the equilibrium issues, the designer had to actively intervene in the growth process by manually checking, relocating, and rotating the positioning of the samples, as seen in Figure 33. This was crucial to prevent any influence from the microclimate, especially since the humidifier was positioned on one side of the chamber while the samples were on the other. Manual interventions were also needed to prevent mushrooms from touching another sample or parts of the chamber during growing. In short, the designer had to closely monitor the sample growth and make manual micro-adjustments, as needed.

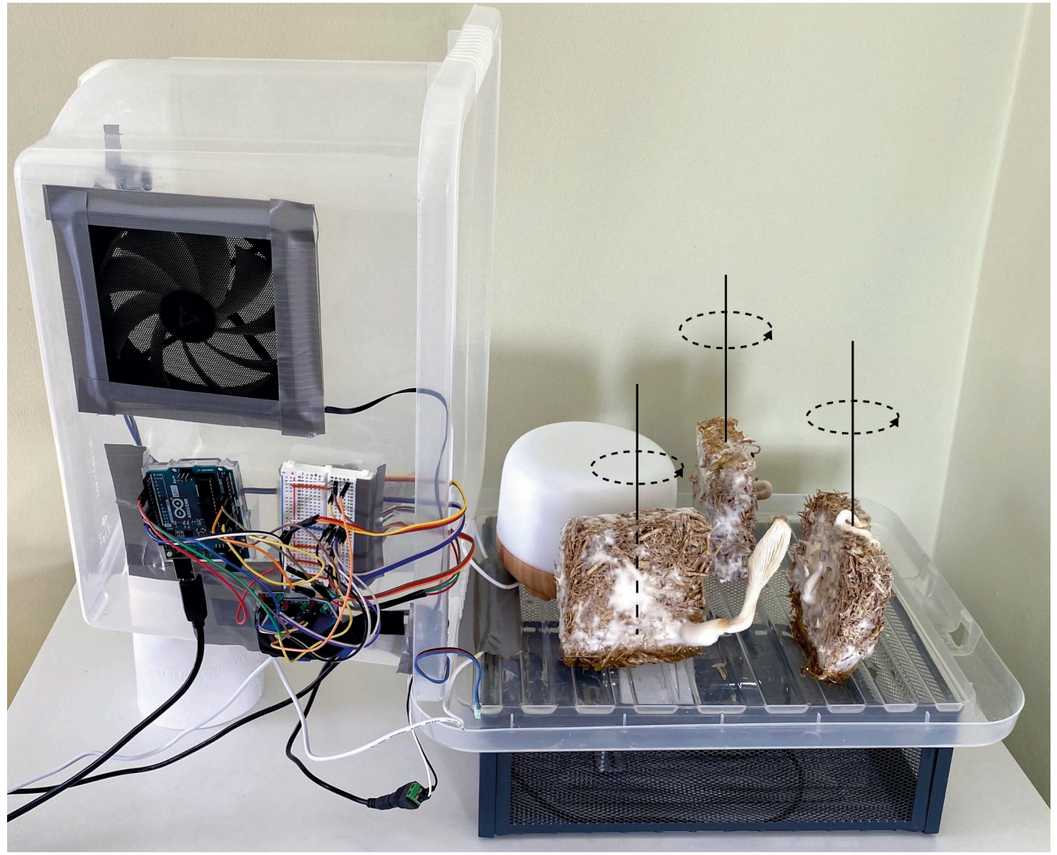


Figure 33

Adjusting the samples in the growth chamber by rotating and moving them.

6. Discussions

6.1. Morphospace

The experiments showed that it is possible to identify specific environmental parameters that influence the morphological space, also known as morphospace, in mushroom development. Morphospace serves as a structured method for mapping, illustrating the diversity of morphological forms based on quantitative traits (Hallgrimsson et al. 2015). The experiments effectively demonstrated how shapes are defined by these quantitative traits. This gave rise to two implications. Firstly, various factors or variables may influence the mushrooms in a similar way. For example, both high humidity and substrate amount will affect the mushroom size. Secondly, a particular variable may be associated with or influence multiple outcomes. For example, humidity affects not only mushroom size but also curvature. This suggests a complex relationship between the variable and the various effects it may contribute to.

Despite these implications, it is still possible to talk about the dominant effects of a particular factor on mushroom morphology. Each variable has a more dominant effect on the growth and development of mushrooms. Based on the experiments, as seen in Figure 34, the morphological feature of mushrooms depends on:

- Humidity: high humidity can cause the caps and stalks to become more curved, while low humidity can cause them to flatten out.
- CO_2 levels: High levels of CO_2 can cause the caps and stalks of mushrooms to become smaller, while low levels of CO_2 can have the opposite effect.
- Gravity: The position of the mushroom samples can affect the bend angle of the stalks. For example, if the samples are vertical, the stalks may be straighter, while if they are horizontal, the stalks may be more curved.
- Substrate amount: The amount of substrate available to the mushrooms can affect their overall size (including both cap and stalk size) and the number of sprouts. More substrates may result in bigger and more numerous mushrooms, while less substrate may result in smaller and fewer mushrooms. Therefore, there is a need for the coefficient value that arises from the proportion of substrate quantity and mushroom size.

It is important to note that these are just a few parameters, and many other factors can influence the growth and development of mushrooms. For example, temperature, light levels, and nutrient content can all affect the morphology of mushrooms.

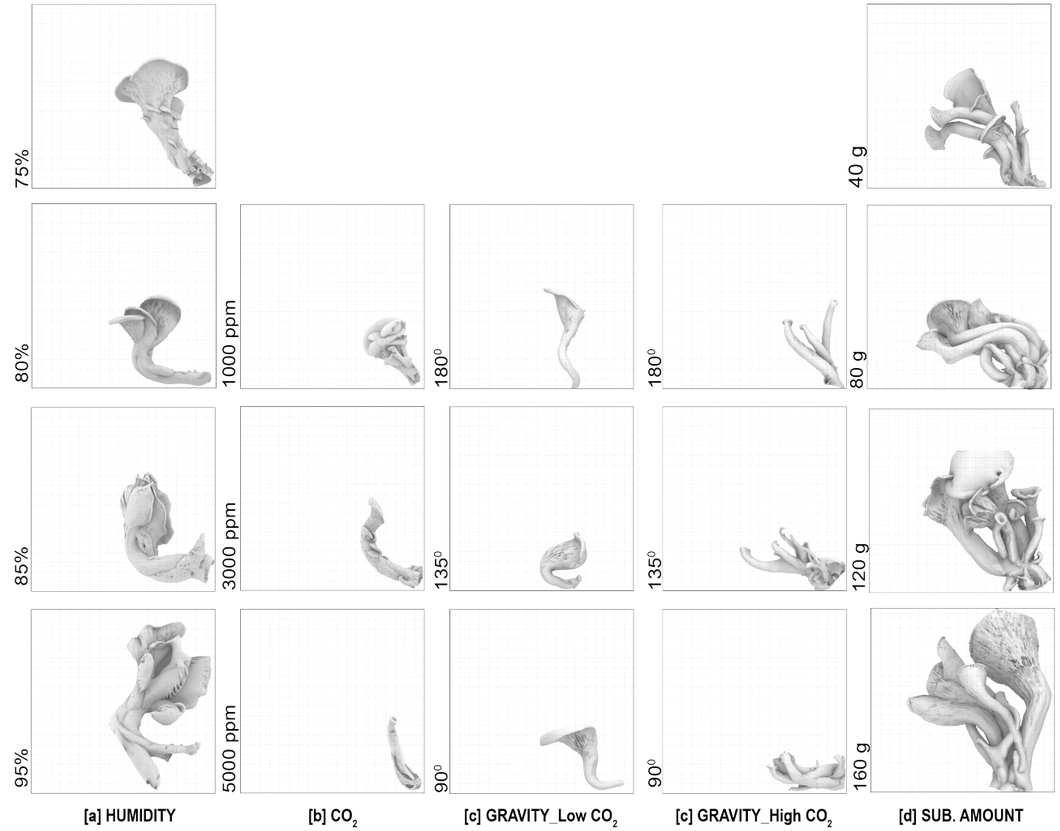


Figure 34

The diagram prepared in Rhinoceros that overlays the 3D scans of the mushrooms (at the same scale) grown under the same environmental conditions: The average sizes and curvatures of mushrooms in the (a) humidity (b) CO_2; (c) gravity; (d) substrate amount experiment.

6.2. Dominant Factor

The experiments revealed a consistent connection between environmental conditions and mushroom characteristics. Cap and stalk curvature are associated with humidity, cap size correlates with CO_2, stalk bend is related to gravity, and overall mushroom size is linked to substrate amount. This means that a single factor can affect more than one feature. As seen in Figure 35, humidity impacts both stalk and cap curvature, while gravity also affects stalk curvature. This causes a complication for a designer aiming to design their morphospace.

To be able to design their morphospace, it is necessary to predict a living material's behaviour, as it helps in understanding the formation of morphological variations. When designing mushrooms based on predictions, it is crucial to focus on a single dominant factor rather than considering either the impact of one variable on various parts or multiple variables that affect a single part in morphology. Meaning, that if a single input impacts two outputs, the one with a stronger correlation will be acknowledged as the dominant factor. For example, in the case of humidity, since the means of error in stalk curvature are larger than those in cap curvature, humidity is identified as the dominant factor for cap curvature. As a result, humidity is the dominant factor for cap curvature, CO_2 for cap size, gravity with stalk bend, and substrate amount with stalk size.

The predictability of mushroom morphologies raises the question of their linearity. While it is acknowledged that living systems are inherently nonlinear, we are still able to foresee their behaviour. Therefore, there is a need to understand the specific nature of their nonlinear system.

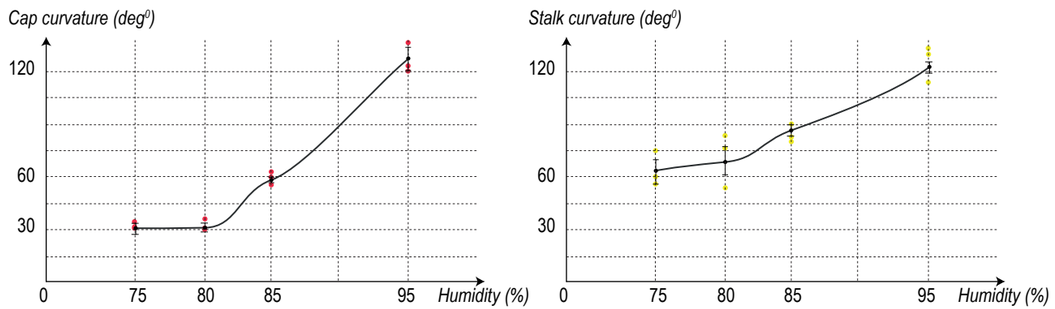


Figure 35

This graph illustrates the correlation between humidity and the curvature in mushroom caps and stalks.

6.3. Nonlinear Correlation

Based on the definition of linearity (Same cause, Same effect, always), the behaviour of mushrooms falls under the category of a nonlinear relationship because they cannot be fully explained using a mechanical, cause-and-effect model. Instead, living systems operate based on an inherent causality that is not straightforward, but involves a continuous process of assessment allowing them to adopt and incorporate uncertainties into their functioning (Zukauskaite 2023). However, it's important to note that not all nonlinear systems are inherently unpredictable. Some exhibit chaos, characterised by noise and tipping points, making them incredibly unpredictable. On the other hand, certain nonlinear systems, like those defined by exponential functions in mathematical formulas, can be predictable. In the case of mushrooms, they demonstrate a correlation between inputs and outputs, enabling a certain degree of predictability in their morphology. However, predicting morphology comes with a margin of error, given the variability in behaviours observed in experimental samples. With only three samples used in the experiments, error bars could impact our understanding of the level of predictability. Nevertheless, despite the potential for high error, predictability is still achievable due to the presence of a correlation.

The design experiments in this chapter demonstrated that fungi in mushroom form exhibit nonlinear properties with both weak and strong correlations. A change in some of the parameters exhibits a distinct trend in one morphological feature. As shown in Figure 36, by looking at the graphs, one can discern the correlated input-output variables. The size or degree of the organism tends to alter with an increase in the input variable. The illustration of the dominant variable that significantly influences a singular change helps to detect the correlation. A high error bar indicates a weaker correlation, as seen in the graphs on the right column. Although the margin of error was not excessively high in all experiments, this problem can be solved by increasing the sample size (to more than three biggest mushrooms) for each experiment. Finding out the correlations and dominant factors is significant from a design perspective. If a variable demonstrates no correlation with morphology, it is not deemed the main (dominant) factor suitable to be used as a variable in a parametric system.

After clarifying that mushrooms exhibit nonlinear behaviour with correlations for some of the inputs, the next question arises: Is this correlation valid at all times? Since the graphs in Figure 36 do not exhibit a straight line, there must be specific start and end points where the correlation exists.

6.4. Thresholds and Tipping Points

A (critical) threshold is a specific point used to characterise a shift in the behaviour in a condition or process ("Thresholds and Tipping Points" 2023). A tipping point is similar, but more closely associated with complex systems, indicating a critical point where the system undergoes a radical and potentially irreversible shift into a different equilibrium state ("Thresholds and Tipping Points" 2023). For example, an elastic material becomes plastic due to an irreversible deformation (DeLanda 2015). Mycelium forming a mushroom can be a tipping point for fungi.

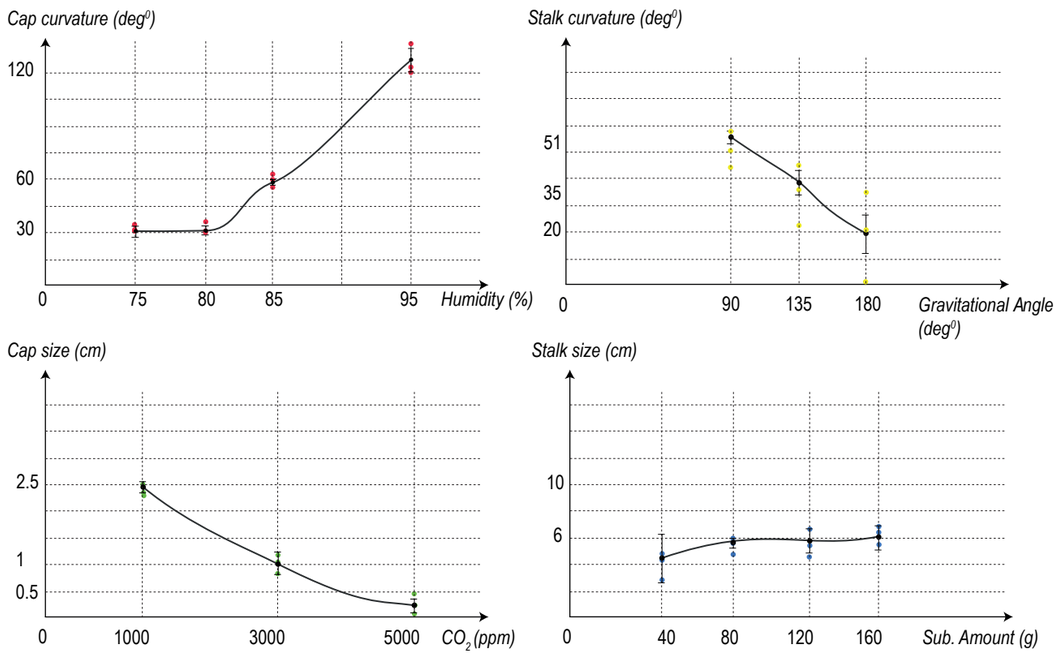


Figure 36

The graph illustrates the correlation between the environmental variable and the variation in mushrooms (left), along with the singular dominant factor that significantly influences specific mushroom parts (right).

While conducting the mushroom experiments, the specific boundaries were set within which the conditions for mushroom life were defined. Therefore, the experiments did not explore the tipping points specifically. However, as we know there are limits for mushroom life, and it is hypothesised that there are points where the growth may change. After these points, there might be drop-offs or significant changes. In mushroom experiments, growth initiation occurs upon meeting the criteria of lower thresholds and continues at a consistent rate until the upper threshold is reached. For example, no growth may be observed at extremely low humidity levels (I could not grow mushrooms under 75% humidity). However, with an increase in humidity, mushrooms undergo a significant behaviour change and grow rapidly. After reaching the upper limit, growth might stop or proceed with a different acceleration rate (which has not been tested in this research). Therefore, it is possible to hypothesise that mushrooms exhibit correlative, nonlinear, parametric properties only within the range defined by the upper and lower critical thresholds. As seen in Figure 37, the line between upper and lower thresholds (in the humidity experiment) is predictable. After reaching the upper threshold, the behaviour of the growth pattern may change, which makes it unpredictable. As a result, understanding the critical thresholds of materials is important for the design processes.

On the other hand, in some cases, material behaviour may be limited, even if the conditions change dramatically. This limitation can be due to various factors, including an organism's genetic code or other growth constraints associated with physical interactions within the organism, such as diffusion limits. For example, mushrooms have a distinct growth pattern and can only grow to a specific size, regardless of the quantity of nutrients available. An oyster mushroom cap, for example, may not reach a maximum of five meters, even when exposed to ample nutrients, reduced CO_2 levels, optimal humidity, and other necessary adjustments.

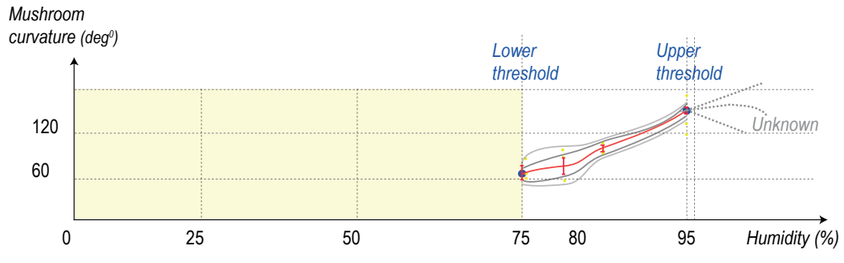


Figure 37

The tipping points on this graph reflect the outcome of my experiments, highlighting the challenges in cultivating oyster mushrooms when humidity conditions do not exceed 75%. The red line represents the actual results of the experiment, while the grey lines illustrate potential variations in the humidity-curvature relationship. Beyond the upper limit, the formation of the line becomes unknown.

Due to the limited time of this research, the experiments could not explore (i) the critical thresholds and tipping points of each variable where growth is inhibited; (ii) tipping points or edges which lead the developmental pathway for the mushrooms to change; or (iii) where normal development is critically disrupted because changing the variable no longer affects (or affects, as expected) the mushroom morphology.

6.5. The Interplay Between Deterministic Processes and Biological Agency

The process of designing with living materials, as seen in the experiments, involves both deterministic and indeterministic processes, balancing control and emergence. Kokotovich defines determinism as the presence of cause-effect relationships governed by physical principles, enabling relatively precise prediction and calculation of outcomes (2012). In contrast, indeterminism refers to the potential for change and the unpredictability inherent in a system (Kokotovich 2012). By this definition, designing with living materials is deterministic because initial growth conditions and parameters can be carefully controlled to guide the system toward intended outcomes. These cause-effect relationships underpin its deterministic nature. At the same time, it is indeterministic due to the agency of living materials. Their biological complexity and (correlational) non-linearity promote emergent behaviours, contributing to unpredictability.

7. Conclusion

This chapter explored the question: Can the growth of mushroom fruiting bodies be guided by environmental factors that influence their developmental plasticity? If this is indeed possible, can we design mushrooms parametrically? To check whether mushroom morphology qualifies as parametric, the chapter distinguished variables in terms of their cause-and-effect relationship. Subsequently, a digitally controlled growth chamber was used to manipulate environmental conditions for design experiments. The chamber helped the digitisation of actuation in the growth and self-assembly processes of a biological system. It provided a potential platform for communication between the designer (via code) and the organism (via morphological response), wherein interactions occurred through continuous modifications of various variables.

The design experiments have demonstrated that the morphospace of mushrooms can be crafted parametrically by manipulating specific environmental parameters. The morphological outcomes of mushrooms exhibited a nonlinear correlation with input parameters such as humidity, CO_2 levels, substrate amount ,and gravitational force, making their development somewhat predictable. However, it is crucial to acknowledge three caveats: (i) a significant margin of error exists; (ii) threshold values apply limits on this bio-digital fabrication method; and (iii) input variables that are linked may affect the same morphological features and adds a complexity to the method. It is also worth noting that the small sample size of the experiments was restricted to a single family of edible mushrooms with only three replicates.

As we understand, the morphospace of mushrooms is predictable within its limitations. A further question arises: Can a design process be developed which would allow a prediction concerning the shape of a mushroom for conditions that have not yet been tested? Can a predictive computational model of mushroom morphology, based on the dominant relationships observed between environment and growth under various environmental conditions, be used as a fabrication method? Finding the conclusions to these questions would offer a design opportunity for nonlinear materials by presenting the parametrisation of a complex growth process leading to complex 3D forms. It will also address challenges arising from the inherent uncertainty of outcomes in the design of biological systems that can be applied to any other form-giving organisms.

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CHAPTER 7

PROBABILITY SPACE

1. Introduction

Chapter 6 demonstrated that mushrooms, when exposed to various environmental conditions, exhibit a nonlinear correlation with consistent morphological patterns of growth and development. The design experiments introduced the idea that mushrooms can be parametrically fabricated within a specific range, lying in between their tipping points. This chapter questions whether it is possible to use the findings from Chapter 6 to develop a predictive computational model of mushroom morphology under varying environmental conditions. Answering this question involves presenting methods for designing living materials under previously untested conditions. It is important to note that the purpose of this investigation is not to construct a catalogue of mushroom shapes, but rather to assess the feasibility of using a parametric approach in a predictive manner.

To predict the morphospace of a mushroom under untested conditions, this chapter initially identifies the dominant factors, that are responsible for a particular morphological feature change. The sizes, curvatures, and overall form predictions are illustrated through 2D development diagrams, based on the outcomes from a focused range of scientific experiments. Subsequently, these drawings were compared with the observed physical growth of mushrooms. The comparison of the diagrams of mushroom morphologies with the actual results showed that morphologies can be predicted within a certain range. This range is referred to as a “space of probability” (or a probability space). It relies on the predictability of an organism’s developmental plasticity.

The design experiments conducted in this chapter aimed to contribute to the understanding of how simultaneous alterations of multiple variables can influence the growth process, potentially resulting in a complex 3D form. The analysis of these experiments helped to demonstrate the viability of using a probability space as a method. Despite the significant variability inherent in mushroom morphology due to its emergent nature and the complex processes underlying it, this method could also be applied to other form-giving organisms.

2. Methods

2.1. Using Data from Previous Experiments

The objective of the design experiments was to create a reliable and formal method which allows the designer to predict the form of their complex living material for conditions that have not yet been tested. Specifically, the goal was to alter more than one variable at a time and see if it was possible to forecast the morphospace of the newly grown mushrooms. The experimental process involved three stages: (i) creating drawings of the expected mushroom morphologies, under predetermined environmental conditions, based on the outcomes of previous experiments in Chapter 6; (ii) growing the mushrooms under the specified environmental conditions; and (iii) comparing the drawn expectations with the actual mushrooms that were grown.

As already known, the growth and development of form in biological systems are often the result of complex interactions between multiple variables rather than the influence of a single dominant factor. Instead of trying to pinpoint the effects of a single input, the experiments in this chapter take a more general approach to test the idea that mushroom morphologies can be predicted. Predictions were formulated based on the nonlinear correlations observed previously. As seen in Chapter 6, **humidity exhibits a strong correlation with cap curvature, CO_2 correlates with cap size, gravitational forces correlate with stalk bend angle, and substrate amount correlates with the stalk size** (Figure 1). Building upon these correlations, I used them to predict the characteristics of newly grown mushrooms.

Three experiments (named 2D, 3D, and 4D) were conducted to investigate the possibility of simultaneously altering multiple variables. The experiments involved changing two, three, and four variables, respectively. All experiments were conducted in triplicate. The setup, including the use of the growth chamber, substrate type, overall duration, temperature, light exposure duration, and methods for recording results, remained consistent with Chapter 6, Section 3. Following an initial three weeks of growth under the same conditions, the samples were exposed to different environmental conditions for eight days within the growth chambers. Through incremental adjustments of more than one parameter across different experiments, as outlined in Table 1, the primary goal was to measure the cumulative effects of different conditions.

The experimental conditions were selected from parameters that had not been previously tested, with the only criterion being to set them within ranges that had been previously tested. For instance, CO_2 levels of 4000 ppm had not been tested specifically previously, whereas CO_2 levels of 1000, 3000 and 5000 ppm had been. In those new and as yet unknown conditions, it was expected that a gradual change in the morphology would occur based on the input values. This was due to the nonlinear correlational behaviour of mushrooms. Mushroom sizes and curvatures in those conditions were calculated assuming they are directly proportionate to the environmental variables.

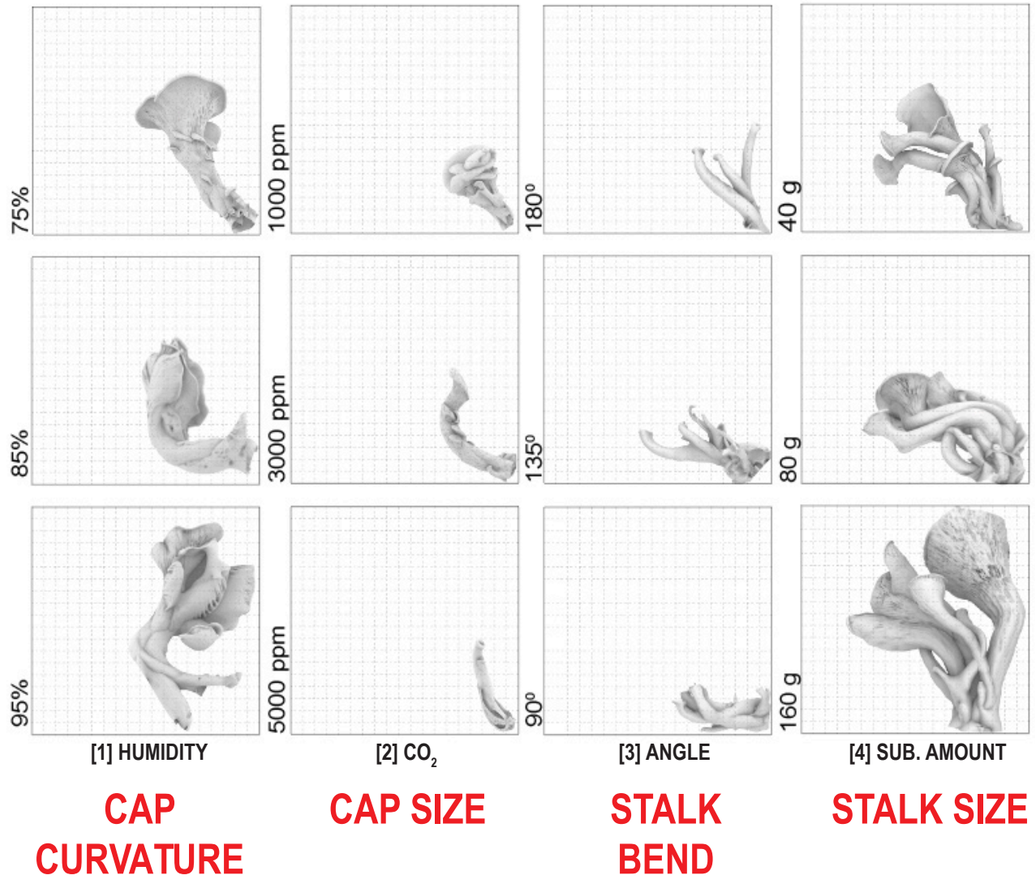


Figure 1

Overlayed 3D mushroom scans illustrating the impact of a dominant factor on morphology. This comparison reveals the following correlations between inputs and outputs: humidity-cap curvature, CO_2-cap size, gravitational force-stalk bend, and substrate amount-stalk size.

Table 1

This table presents the list of variables employed in experiments [2D], [3D], and [4D].

Variables	Experiments		
	[2D]	**[3D]**	**[4D]**
Humidity (%)	95	75	95
CO_2 (ppm)	4000	1000	2000
Substrate (g)	55	55	80
Gravity (deg.^{0})	90^0	180^0	45^0 and 180^0
Light (nm)	4h, 450 nm	4h, 450 nm	4h, 450 nm
Temp. (0^C)	20-22^{0}C	20-22^{0}C	20-22^{0}C

2.2. Drawing of Expectations

While predicting the characteristics of newly grown mushrooms under specific conditions, the data on morphological features was retrieved from the previous experiments based on the dominant factor or the input that exhibits a strong correlation. This data, specifically regarding morphological features, can be found in Chapter 6, Tables 3a, 4a, 5a, and 6a. Each characteristic of the newly grown mushrooms corresponded to a specific experimental condition:

- Cap curvature from the humidity experiment,
- Cap size from the CO_2 experiment,
- Stalk bend angle from the gravity experiment,
- Stalk length data from the substrate amount experiment.

Using this information, as summarised in Table 2, drawings of expected mushroom morphologies were drafted before conducting the experiments, without anticipating the results. For each set of experiments, minimum and maximum values, signifying the range across three samples, were selected to represent the extremes. Furthermore, average values were computed from the data gathered from each of the three samples. These illustrative drawings, based on Table 3, served as visual representations of the anticipated results. They were intended to provide insight into stalk and cap size and curvature. Their purpose was to help designers predict potential morphological outcomes and plan the experiment accordingly for future work.

Based on the dimensions provided in Table 3, two-dimensional drawings were generated using Adobe Illustrator and AutoCAD. Initially, a 0.5x0.5 cm grid was established. Subsequently, five potential variations, for each condition, were illustrated within this grid. The smallest and largest sizes or degrees of curvature of caps and stalks were depicted in grey using lighter and dashed lines to indicate a lower likelihood of mushroom growth in those configurations. Conversely, average mean values were represented by a dark and thick line, indicating a higher likelihood of mushrooms growing in those forms, as illustrated in Figure 2.

Table 2

Summary of the data retrieved from Chapter 6, (Tables 3a, 4a, 5a, and 6a) presenting the dominant relationships between environment and growth. Minimum and maximum values, as well as average measurements, were obtained from three samples.

	Humidity *For 2000 ppm, 55 g substrate and 90^0 angle*	**95%**	**85%**	**80%**	**75%**
Cap	Min curvature (^{0})	127^0	53^0	30^0	31^0
	Max curvature (^{0})	145^0	60^0	38^0	36^0
	Average curvature (^{0})	130.7^0	59^0	33^0	33^0

	CO_2 *For 80% humidity, 55 g substrate and 90^0 angle*	**5000 ppm**	**3000 ppm**	**1000 ppm**
Cap	Min size (cm)	0.1	0.8	2.3
	Max size (cm)	0.4	1.3	2.5
Stalk	Averagesize (cm)	0.2	1	2.4
	Average length (cm)	2.3	4.2	2

	Gravity *For 75% humidity, 55 g substrate and 5000 ppm*	**0-90^0**	**45-135^0**	**0-180^0**
Stalk	Min curvature (^{0})	44^0	23^0	0^0
	Max curvature (^{0})	57^0	46^0	30^0
	Average curvature (^{0})	50.7^0	34.7^0	19.7^0

	Substrate amount *For 80% humidity, 90^0 angle and 3000 ppm*	**40 g**	**80 g**	**120 g**	**160 g**
Stalk	Min length (cm)	2.9	5.2	5.2	5.4
	Max length (cm)	5.3	6	6.7	6.7
	Average length (cm)	4.4	5.7	5.9	6.1

Table 3

Summary of the expected mushroom sizes and curvatures based on Table 3.

Variables	Experiments		
	[2D]	**[3D]**	**[4D]**
Cap size (cm)	Between 1 - 0.2	Between 2.3 - 2.5	Between 1-2.4
Cap Curvature (^{0})	Between 59^0 - 131^0	Between 33^0 - 0^0	Above 59^0
Stalk size (cm)	Between 2.3- 4.2	Between 2 – 4.2	Between 4.4 - 5.9
Stalk Curvature (^{0})	Between 44^0 - 57^0	Between 30^0 - 0^0	Between 11^0 - 41^0

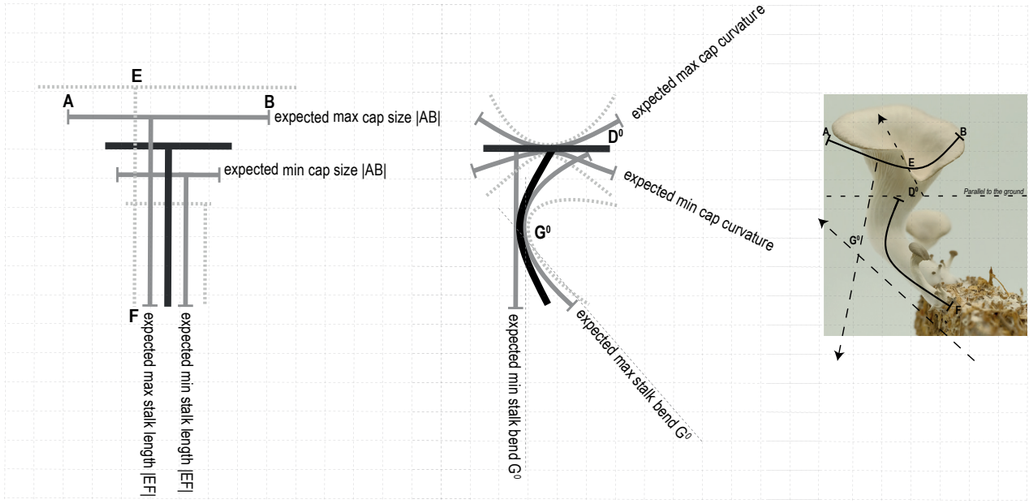


Figure 2

A diagram illustrating the locations of the measurement points in 2D for various morphological features. The grid size used was 0.5x0.5 cm. Lighter, grey, and dashed lines represent a lower likelihood of mushroom growth, whereas darker and thicker lines signify a higher likelihood of mushrooms growing in those forms.

In summary, the steps taken to predict the newly grown mushroom morphologies were:

- **Parameter testing:** The impact of environmental parameters on mushroom growth was tested (conducted in Chapter 6).
- **Dimension recording:** The dimensions of the mushrooms grown within these environmental parameters were recorded. This involved measuring characteristics like cap diameter, stalk length, and overall size (covered in Chapter 6, Tables 3a, 4a, 5a, and 6a).
- **Identification of dominant factor:** Data analysis was conducted to identify the dominant environmental factor correlating with a specific morphological change. For example, it was found that CO_2 had the most significant impact on cap diameter (explored in Chapter 6).
- **Adjustments of new parameter values:** New values were set for the parameters in the experimental setup.
- **Determination of range with lower and higher likelihood:** Data from the previous experiment was used to establish the potential range of dimensions under new environmental conditions. Ratios and proportions derived from past data were employed to estimate lower and higher likelihoods.

  For example, if increasing humidity typically led to a certain percentage increase in cap curvature, the curvature at 85% humidity was expected to fall within the range observed at 75% and 95%. These values represented the lower likelihood as a potential range. To determine the dimension with the higher likelihood, the midpoint between the minimum and maximum values for curvature at 85% humidity was calculated.
- **Visualisation of expected morphologies:** Once the minimum, maximum, and average likelihoods were calculated based on the adjusted parameters, the expected morphologies were visualised. The minimum and maximum dimensions were used to represent the upper and lower sizes of the features (such as cap diameter and stalk length), while the average values represented the more likely dimensions. This step helped the visualization of the range of potential morphologies resulting from the changes in environmental parameters.

I. 2D experiment expectations

Based on the CO_2 experiment in Chapter 6, mushroom caps growing at 4000 ppm were expected to have caps between 1 cm and 0.2 cm. Based on the humidity experiment, the expected cap curvature was approximately 130^0 (or more than 59^0). Based on the gravity experiment, stalk curvature was expected to fall between 44^0 and 57^0.

Although the stalk size typically corresponds to the substrate amount, in specific instances, it was influenced by the CO_2 experiment, since the same amount (55g) of substrate was used.

Consequently, stalks were expected to fall within the range of 2.3 cm to 4.2 cm. Additionally, it is worth noting that high humidity tends to result in a larger volume of mushrooms. The previous CO_2 experiment was conducted at 80% humidity, whereas this one was set at 75%. Therefore, it was expected that the stalks might be closer to the lower limit in the predicted (2.3 to 4.2 cm length) range. However, due to the uncertainty regarding how much shorter the stalks could be, the predictions were left in a wider range. According to these measurements (Table 3), the mushroom morphology is illustrated in Figure 3.

II. 3D experiment expectations

Based on the CO_2 experiment in Chapter 6, mushroom caps growing at 1000 ppm were expected to have caps approximately 2.4 cm and stalks between 2 cm and 4.2 cm. Since the substrate amount was 55g, the stalk was determined based on the CO_2 experiment, rather than the substrate amount experiment.

Based on the humidity experiment, the cap curvature was expected to be around 33^0 (or less). Based on the gravity experiment, stalk curvature was expected to be approximately 19^0 (or less than 36^0). According to these measurements, the mushroom morphology is depicted in Figure 4.

III. 4D experiment expectations

Based on the humidity experiment in Chapter 6, the cap curvature was expected to be approximately 130^0 (or more than 59^0). Based on the CO_2 experiment, mushroom caps grown at 2000 ppm were expected to exhibit caps ranging between 1 cm to 2.4 cm. Based on the substrate amount experiment, stalk length was expected to be approximately 5.7 cm (or between 4.4 and 5.9 cm).

Lastly, with all variables held constant, the direction of the gravitational force was altered after three days. This change was done by adjusting the position of the substrate blocks. Based on these conditions, the initial stalk curvature was expected to be below 36^0. After reaching half-length, the stalk was expected to bend within the range of 23^0 and 46^0. By calculating the minimum and maximum averages of these values, it was estimated that the stalk curvature would fall between 11^0 and 41^0. Based on these measurements, the expected mushroom morphology is depicted in Figure 5.

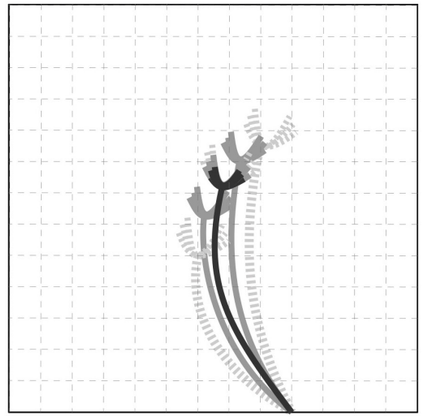


Figure 3

Illustrations of the anticipated mushroom under the following conditions: Humidity 95%, CO^2 4000 ppm, Substrate Amount 55g, Gravity 90^0.

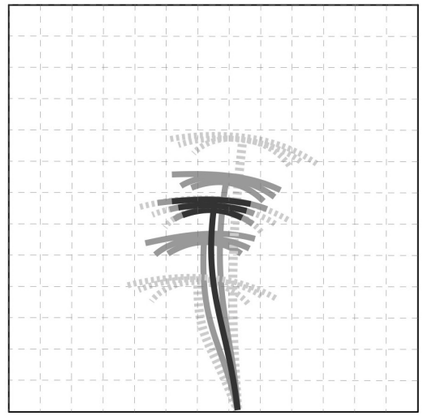


Figure 4

Illustrations of the anticipated mushroom under the following conditions: Humidity 75%, CO_2 1000 ppm, Substrate Amount 55g, Gravity 180^0.

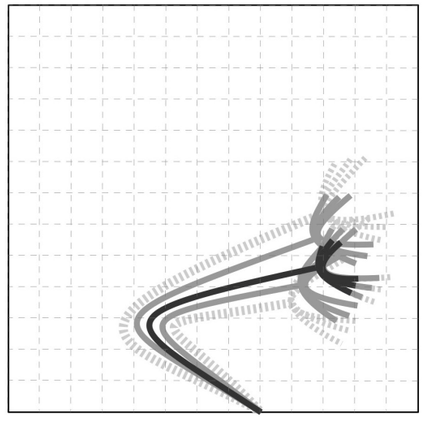


Figure 5

Illustrations of the anticipated mushroom under the following conditions: Humidity 95%, CO_2 2000 ppm, Substrate Amount 80g, Gravity initially 45^0 and then 180^0.

2.3. Physical Experiments

The parameters of the experimental setup are summarised in Table 2. After setting up the parameters, the same growth chamber and methods used in Chapter 6 for measuring, recording the results, and calculating the deviation were employed.

I. 2D experiment [Humidity - Aeration]

The ambient condition was adjusted to 95% air humidity. The other variable, CO_2 level, was set at 4000 ppm. Light exposure was kept constant (4 hours per day at the same time of the day). The substrate amount was 55g, sample positioning was at 90^0, and the temperature was maintained at 22^0C throughout this set of experiments.

II. 3D experiment [Gravity - Humidity- Aeration]

The ambient condition was adjusted to 75% air humidity. CO_2 level was set at 1000 ppm. The samples were positioned at a 180^0 angle. Light exposure was kept constant (4 hours per day, at the same time each day). The substrate amount was 55g, and the temperature was maintained at 22^0C throughout this set of experiments.

III. 4D experiment [Gravity - Substrate amount - Humidity - Aeration]

Except for light exposure (4 hours per day) and temperature (maintained at 22^0C), all the variables were modified. The ambient condition was set to 95% air humidity, CO_2 level at 2000 ppm, substrate amount at 80g, and sample positioning at a 45^0 angle for the initial 3 days. For the last 3 days, the sample positioning was changed to a 180^0 angle from the first position.

2.4. Contrasting Actual Mushroom Growth with Illustrated Expectations

The mushroom formations were compared on a macro scale, helping in the evaluation of the proximity between the expected outcomes and the actual reality. Two approaches were used to see the similarity between expected and actual morphologies. The initial approach involved a visual examination, wherein drawings and overlayed 3D scans were juxtaposed side by side for a direct visual comparison. As in previous experiments, the mushroom morphology was documented on Day 27 using photography and 3D scanning. The 3D scans of mushrooms cultivated under identical environmental conditions were overlayed digitally for comparison without using image comparison software, as differences/similarities between them were obvious to the naked eye.

In addition to the visual comparison, a numerical analysis of the measurements for both expected and actual mushrooms was conducted. This involved calculating measures of central tendency (average values) and measures of dispersion (error of the mean) as mentioned in Chapter 6. Subsequently, these numerical values between the expected and actual results were compared to provide a more objective assessment of the degree of similarity or difference between the two datasets.

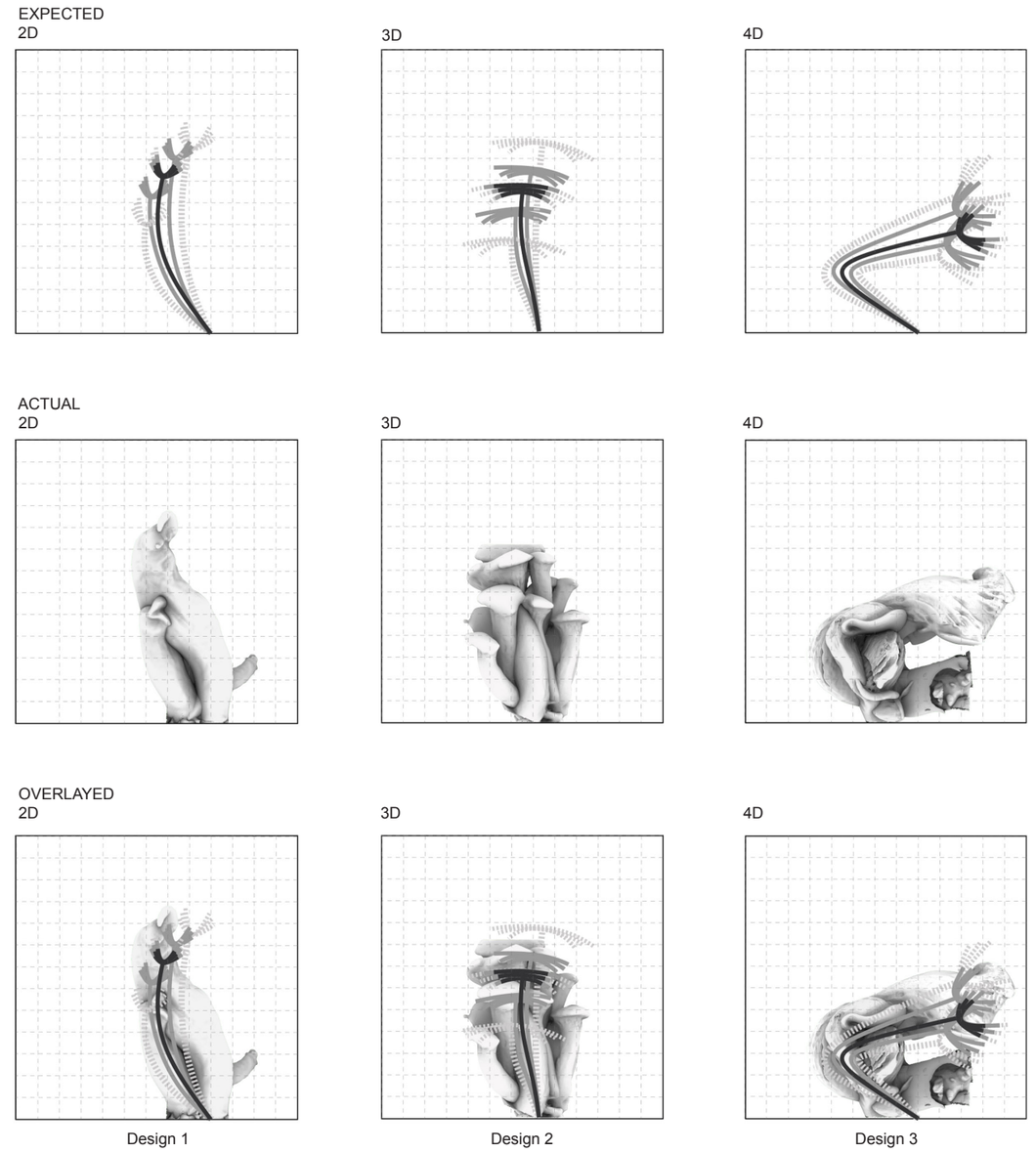


Figure 6

Visual comparison of 2D (Column 1), 3D (Column 2), and 4D (Column 3) experiments. Each row represents drawings, 3D-scanned and overlaid mushrooms, and scanned images overlaid with drawings. The grid size used was 0.5x0.5 cm.

3. Results

The findings indicated the potential to predict the overall morphological tendency or trend in growth patterns based on specific variables (Figure 6). It is crucial to acknowledge that while the predictions were not 100% accurate, they provided insights into how the organism may be influenced by various factors. Moreover, mushrooms growing under identical conditions exhibited morphological similarities.

Figure 7 shows that when influenced by a combination of environmental conditions, newly grown mushrooms exhibit a morphological characteristic reminiscent of previously demonstrated traits. For example, in the first row, the mushroom cap is curved, while in the other picture, the cap size is of medium size. The result (on the right) is a mushroom with a medium-sized, curved cap. This new morphology suggests a form of morphological hybridisation, resulting in a combination of characteristics acquired from the initial experiments.

I. Results of 2D experiment

In Figure 8, morphologies' expectations and reality closely match. Examination of Table 4 reveals that, overall, the newly cultivated mushrooms exhibited the anticipated characteristics. However, that a single sample exhibited deviations in both stalk length and curvature, falling outside the expected range with no growth observed.

The absence of mushroom growth in the third sample led to relying on data from only two samples for the results. This underscores the importance of recognising non-growth as a morphological state deserving attention. In future tests, instances with no growth should be considered, to avoid potential biases, and would potentially increase the standard error. Despite this, the outcomes closely aligned with the predicted values.

II. Results of 3D experiment

As seen in Table 5 and Figure 9, the cultivated mushrooms exhibited the anticipated growth pattern. However, there was a deviation from the expected cap size; the caps were anticipated to be larger but instead appeared smaller, likely due to the adverse impact of low humidity on overall mushroom size. Aside from the smaller caps, the outcomes aligned with the predicted values.

III. Results of the 4D experiment

As seen in Table 6 and Figure 10, the newly grown mushrooms formed as expected. However, there were inaccuracies in the estimated values, particularly in the stalk lengths. Even though it was known that mushrooms growing with 80g substrate would be larger than those with less nutrients (such as 55 g), the extent of this size difference was unknown. However, apart from this limitation, the results were still close to the predicted values.

Figure 7

The photos in the rightmost column show mushrooms displaying certain physical properties, such as cap and stalk curvature, and cap and stalk length, that are shared with those in the preceding columns, suggesting the growth of fungi in hybrid environments.

Table 4

The recorded measurements of actual mushrooms, as well as the expected mushroom size and curvatures, for the 2D experiment. The measurements highlighted in red indicate values that fell outside the expected range.

		Actual Results of [2D]		Expected Results
		Rep-1A	Rep-2A	
Cap	Size (cm)	0.5	0.5	
	Curvature (^{0})	60^0	58^0	
	Average size	0.5		Between 0.2 - 1
	Average curvature	59^0		Between 59^0 - 130^0
Stalk	Length (cm)	3.7	2.2	
	Curvature (^{0})	60^0	57^0	
	Average length	2.7		Between 2.3- 4.2
	Average curvature	58.5^0		Between 44^0 - 57^0

Figure 8

Photographs of two samples as results of the 2D experiment, showcasing left side views (column 1), front views (column 2), right side views (column 3), and detailed close-ups (column 4).

Table 5

The recorded measurements of actual mushrooms, as well as the expected mushroom size and curvatures, for the 3D experiment. The measurements highlighted in red indicate values that fell outside the expected range.

		Actual Results of [3D]			Expected Results
		Rep-1A	Rep-2A	Rep-3A	
Cap	Size (cm)	1.1	1.2	0.8	
	Curvature (^{0})	24^0	0^0	0^0	
	Average size	1			Between 2.3 - 2.5
	Average curvature	8^0			Between 33^0 - 0^0
Stalk	Length (cm)	2.7	2.5	2.5	
	Curvature (^{0})	36^0	18^0	11^0	
	Average length	2.5			Between 2- 4.2
	Average curvature	21^0			Between 36^0 - 0^0

Figure 9

Photographs of three samples as results of the 3D experiment, showcasing left side views (column 1), front views (column 2), right side views (column 3), and detailed close-ups (column 4).

Table 6

The recorded measurements of actual mushrooms, as well as the expected mushroom size and curvatures, for the 4D experiment. The measurements highlighted in red indicate values that fell outside the expected range.

		Actual Results of [4D]		Expected Results
		Rep-1A	Rep-2A	
Cap	Size (cm)	2.2	1.2	
	Curvature (^{0})	139^0	75^0	
	Average size	1.7		Between 1 - 2.4
	Average curvature	107^0		Above 59^0
Stalk	Length (cm)	5.4	2.1	
	Curvature (^{0})	13^0	18^0	
	Average length	3.7		Between 4.4 - 5.9
	Average curvature	15.5^0		Between 11^0 - 41^0

Figure 10

Photographs of two samples as results of the 4D experiment, showcasing left side views (column 1), front views (column 2), right side views (column 3), and detailed close-ups (column 4).

4. Discussions

4.1. A Probability Space

The results showed that correlational nonlinear relationships between the environmental factors and mushroom morphologies are predictable. However, there were significant variations within these correlations. These variations were integral components of the biological parametric relationship. That is why it is more accurate to define potential morphological outputs (or values) as an area, rather than pinpointing them as single points or along a singular line. These areas, where the correlation exists, created a space of probabilities that covered a full range of potential morphological outputs; meaning, the space of probabilities, defined by means of error, recognised that living materials exhibit varying levels of certainty under different factors.

Probability is a mathematical concept concerned with analysing random phenomena or determining the likelihood of events occurring within a defined set of possibilities (Siegmund 2025). It defines a set of possibilities and associates each of them with a probability—a number between 0 and 1 (representing certainty, encompassing all possibilities) (Koller and Friedman 2009). A probability space provides the foundation for this analysis and consists of three key components (Siegmund 2025):

- **The sample space (Ω)** represents the set of all possible outcomes of an experiment. Although the outcome of an event cannot be determined beforehand, it is always one of the possible outcomes defined within the sample space.
- **The sigma-algebra (F)** is a collection of well-defined subsets of the sample space, known as events, which satisfy specific axioms such as closure under union and complementation.
- **The probability measure (P)** is a function that assigns probabilities to these events in the sigma-algebra, adhering to the axioms of probability, including non-negativity, normalisation, and countable additivity.

The concept of probability space goes beyond mathematics and has been applied across various disciplines, such as finance, chemistry, biology, and other fields, to work with uncertainty and randomness, enabling more accurate predictions and decision-making processes (Tijms 2004). It has also proven valuable in architecture, particularly in building design, where it is used to explore possible design options in generative design processes (Wang, Li, and Sun 2025), optimise space allocations (layouts) (Rahbar et al. 2019), and assess building performance attributes (Gerber, Pantazis, and Wang 2017).

Probabilities can be represented using graphical networks, such as Bayesian and Markov networks, to help understand dependencies among variables and analyse complex systems with interconnected elements (Koller and Friedman 2009). However, in this thesis, it was used as an architectural representation method. For the generation of the space of probabilities, 2D drawings were initially used as a visual representation of the designer's predicted morphology for future mushroom growth. These drawings served as the initial step in creating a probability space, offering insight into the potential form of newly grown

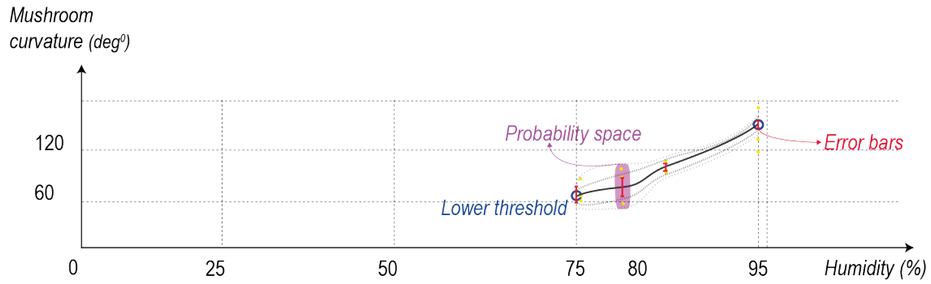


Figure 11

The graph depicting various lines along with their range, illustrating the curvature of mushrooms at 80% humidity, generates a probability space.

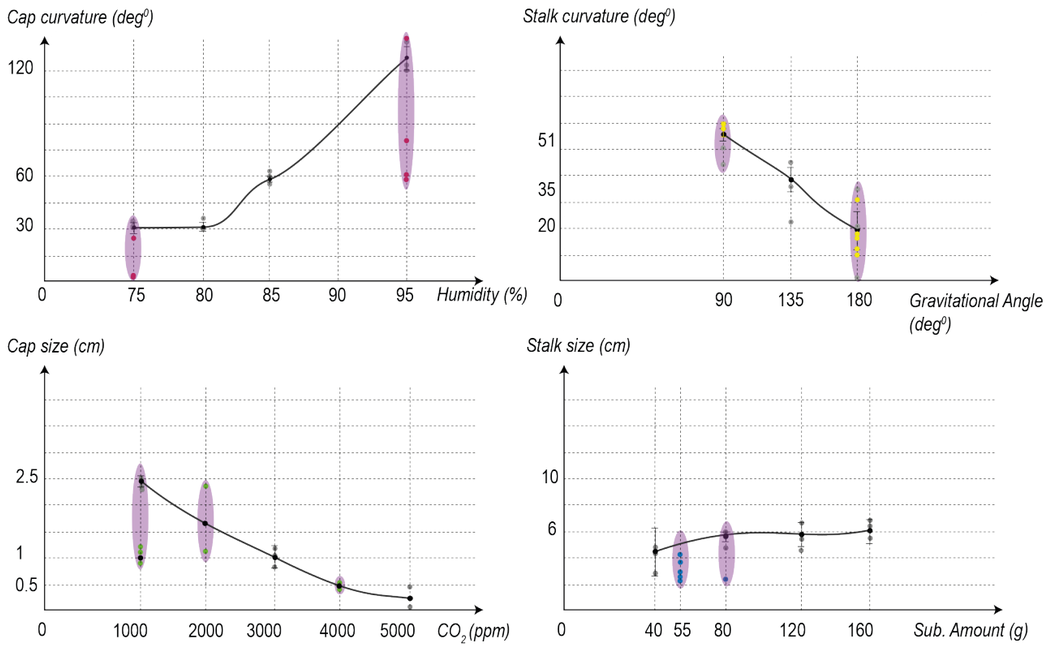


Figure 12

Graphs illustrating the varying levels of certainty by the newly grown mushrooms.

mushrooms. However, these 2D drawings were flat and did not define a space without the third dimension. To address this limitation, a "**probability space method**" was proposed. This probability space method was less predictions of form than predictions of the distribution of matter. It encompassed a set of possible forms in the third dimension, providing a more comprehensive understanding of mushroom morphology. It served as a conceptual tool to describe the chance of different outcomes occurring. The proposed probability space method was a designerly representation of a specific area - a spatial range wherein newly grown mushrooms could form.

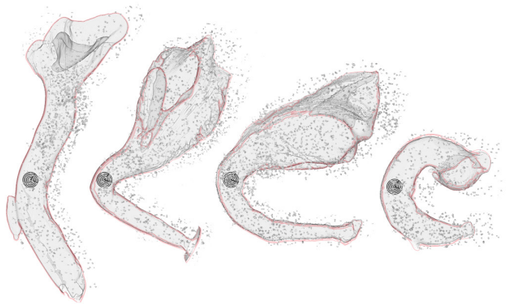
The probability space in this thesis builds upon an understanding of the sample space, which represents all possible oyster mushroom morphologies, capturing the full spectrum of phenotypical variations under diverse environmental conditions. This sample space establishes the morphological boundaries within which oyster mushrooms can grow. Within this framework, the sigma-algebra refers to specific subsets of morphologies that emerge under a given set of environmental conditions, representing a focused range of possibilities within the broader sample space. A particle-based system is used as the probability measure, illustrating the likelihood of new mushroom growth patterns under particular environmental conditions and offering a dynamic representation of spatial and morphological probabilities.

The probability space is generated using a particle-based system, by using calculations from previous experiments involving grown mushrooms. It identifies varying growth possibilities for fungi, distinguishing between areas with low and high growth likelihoods. The concept of a probability space might be reminiscent of the concept of tolerance; however, it represents a spectrum of morphologies rather than various levels of tolerance. The interpretation of tolerance varies across fields such as biology, engineering, material science, or manufacturing. In engineering and biology, tolerance is used to determine the amount of space required for error or the capacity for survival (Oxford Languages). It reflects an organism's ability to survive in high and low levels of environmental factors without adverse reactions. In this thesis, the extremities of high and low tolerance values differ from upper and lower thresholds. Within these thresholds, an organism remains alive, but may undergo behavioural changes. In contrast, tolerance represents a broader spectrum of survival for the organism. Therefore, the probability space does not represent the range of tolerance; instead, it only represents the potential states of the organism under specific environmental conditions. The probability space does not necessarily include tipping points or high and low thresholds (Figure 11).

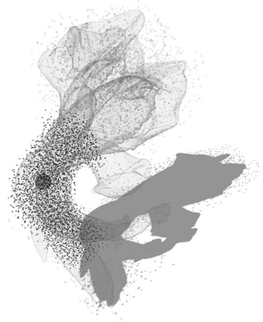
As illustrated in Figure 12, mushrooms exhibit distinctive growth patterns under identical environmental conditions. Although they exhibit a general morphological trend, each mushroom differs from the others, and it is impossible to have identical sizes and curviness.
The probability space drawings represent the diverse 3D forms that mushrooms may take under a specific set of environmental conditions. In this study, the model was developed using an average of two and three mushroom samples. However, introducing more samples could make the probability space more definitive.

1. Assigning 3D scanned mushrooms as nCloth's

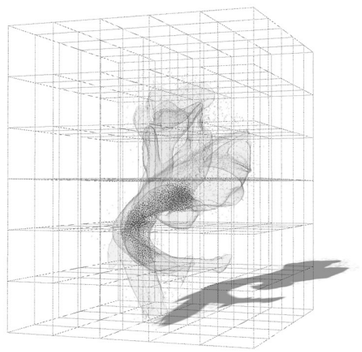
2. Creating passive colliders within the nCloth



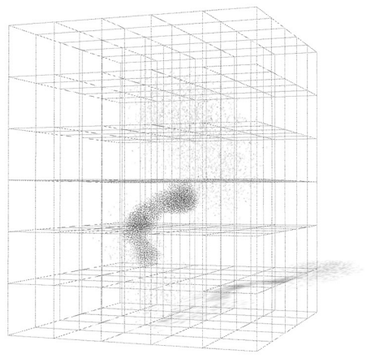
3. Generating nParticles within the nCloth



4. Overlaying nParticles that create the *probability space,* with denser areas highlighted through darkening.



5. Placing the probability space within a matrix cube composed of 150 mini cubes



6. Probability space particles within the matrix cube

Figure 13

A diagram illustrating the steps involved in creating a probability space using Maya.

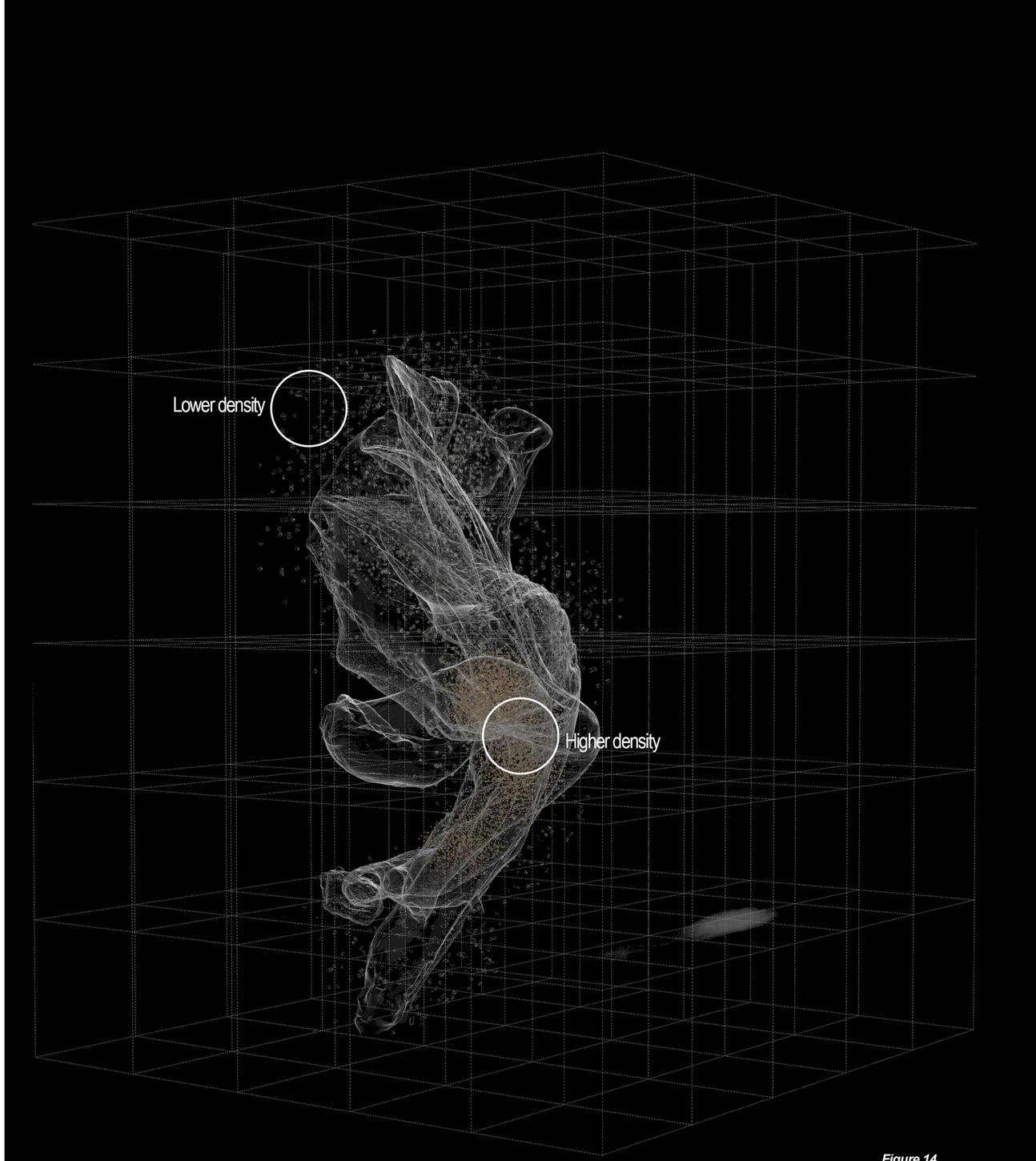
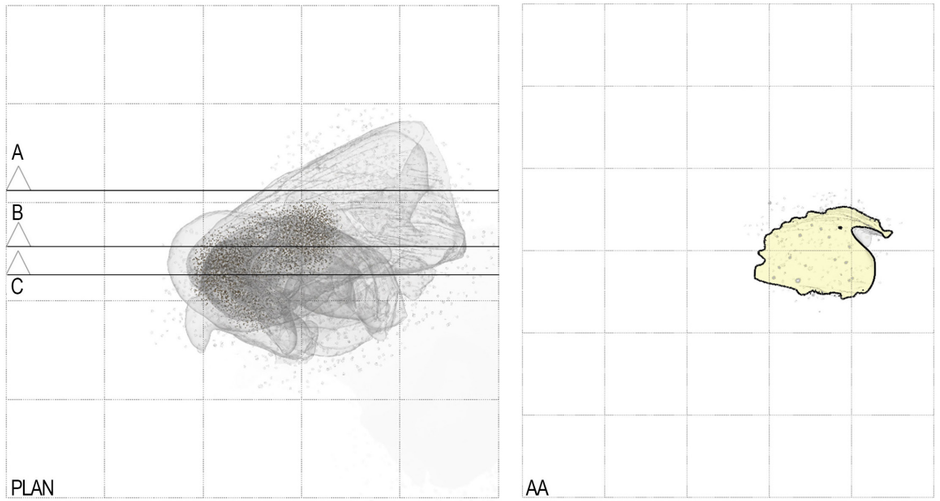


Figure 14

A probability space illustrating the likelihood of a newly grown mushroom being in a specific area.



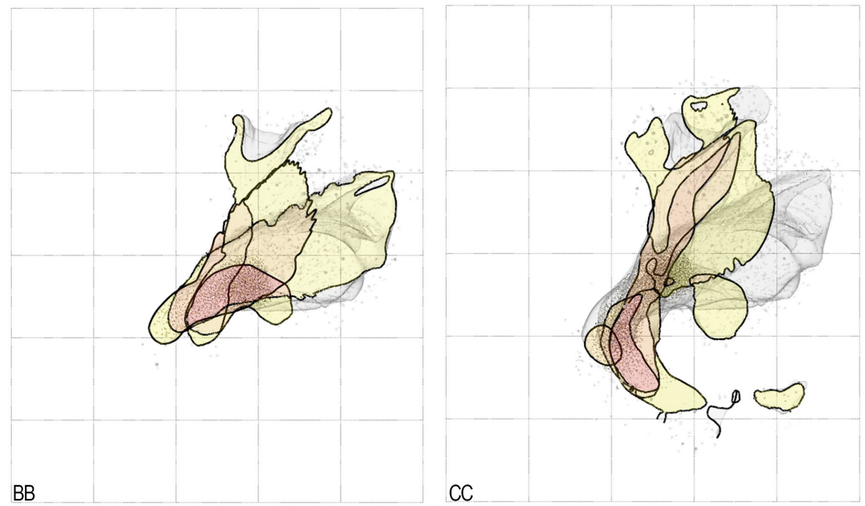
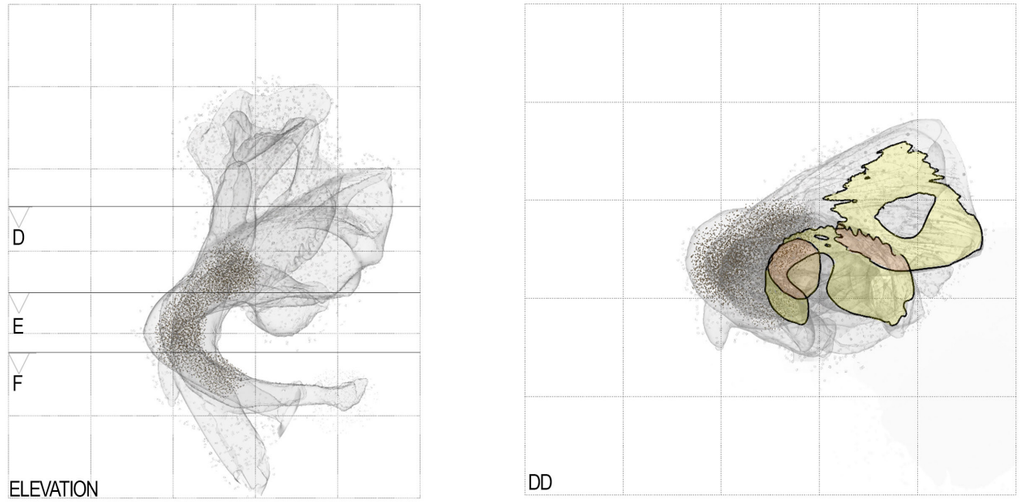


Figure 15

Section drawing of probability spaces for mushrooms grown under 95% humidity. The top-left drawing indicates where the section was cut. AA, BB, and CC illustrate the section cuts of the probability space. Areas with denser particles or darker colours represent higher probabilities.



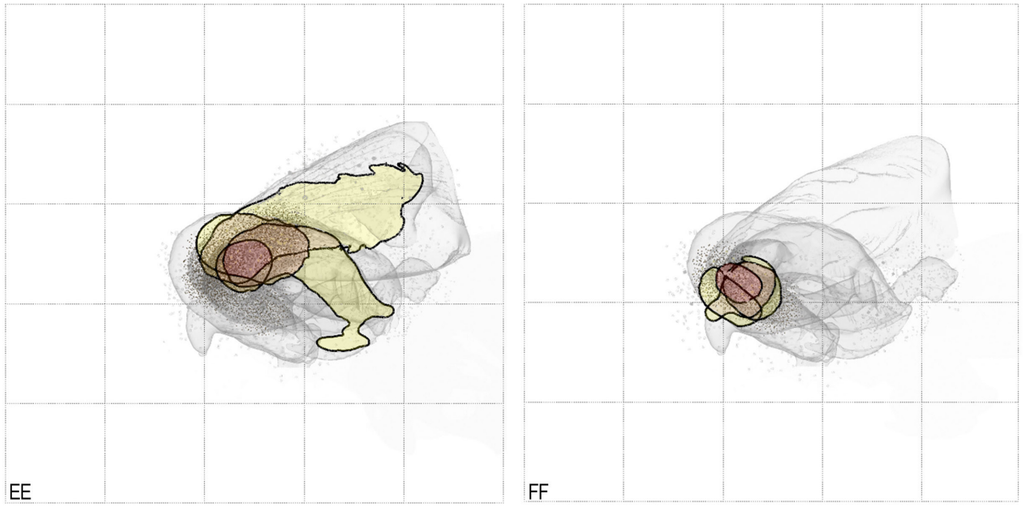


Figure 16

Plan drawing probability spaces for mushrooms grown under 95% humidity. The top-left drawing indicates the levels of the plan. DD, EE, and FF represent the plans within the probability space. Areas with denser particles or darker colours represent higher probabilities.

The goal of predicting through probability space drawings was to enhance the designer's ability to engage with the agency of living materials during the design processes. This approach reflected the collaboration between human intention and material agency, informed by both physical and digital explorations to improve predictability. Mushrooms played a central role in this process and provided valuable data and insights; understanding their non-linearity was crucial. The targeted design outcomes (or artifacts) were mushrooms with specific morphologies, serving as proxies to test whether a designer can predict the behaviours, forms, properties, or functions of a material artifact that simultaneously acts as both a fabricating agent and the material itself.

Predictions of mushroom morphologies under untested conditions were intended to guide designers in adjusting input variables and optimizing growth conditions to achieve desired outcomes. In this thesis, CAD tools, such as Maya, were employed to support designers in making these predictions rather than replacing physical experimentations. However, these tools required integration with data from proxy materials, as they alone could not generate accurate predictions independently.

Maya, a 3D modelling, animation, and simulation tool, was primarily introduced to create probability space drawings. It facilitated the mapping and analysis of the phase space of mushroom growth. Any software capable of generating a particle system and dynamically interacting with other geometries—such as previously 3D-scanned mushroom models—could be used to inform the process. This integration of digital and material approaches provided a robust framework for navigating the complexities of designing with living materials.

A probability space consists of a base and particles. The process to create the particles for these experiments is akin to generating small balls within an inflated balloon. Initially, mushrooms grown under the same environmental conditions were 3D scanned and overlayed using Rhinoceros, creating a foundational representation for each condition. As the next step, a particle system interacting within the volume was built using Maya tools, including nCloth, passive collider, and nParticles (Figure 13). The use of **nCloth** helped the simulation of a diverse range of dynamic polygon surfaces, linking them to the particle system. The polygon surfaces, created from the 3D scanned mushrooms, are assigned as nCloth. To make nCloth interactive, a passive collider was assigned to it. The **passive collider** helped to control the formation and location of particles by guiding their aggregation. Lastly, **nParticles** were generated to depict the uncertainty and indeterminacy inherent in the growth process. These particles helped to maintain ambiguous and cloudy boundaries, avoiding sharp, deterministic borders that could inaccurately represent the unpredictable aspect of living materials' growth. nParticles were generated within the boundaries of the nCloth, by assigning it as a passive collider. They were distributed evenly within each volume delineated by the 3D scans, and overlapping volumes were therefore represented with higher densities. In summary, the transformation of mushroom scans into nCloth first, and a passive collider after, enables the particles to organise around the base of 3D scans of mushrooms. Finally, the base and particles were enclosed within a matrix cube, consisting of 150 mini cubes with dimensions of 0.2x0.2x0.2 cm each. This configuration allowed for a more precise observa-

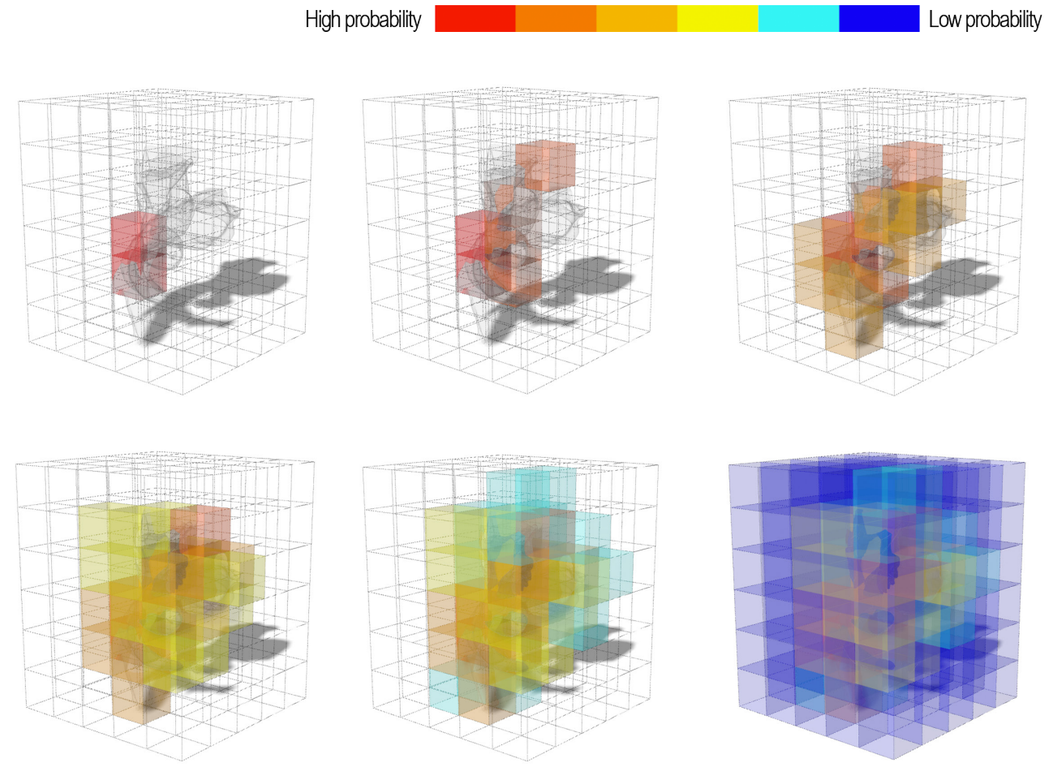


Figure 17

The illustration of the proposed 3D model involves populating a matrix with varying probabilities, indicating the likelihood of a pixel containing the organism—some pixels exhibiting a high chance while others a low chance.

tion and comparison of mushroom growth in the third dimension.

The properties of particles including viscidity, size, propagation speed, and distance from the nCloth surface, could be adjusted independently from the passive collider. These adjustments were made to visually distinguish between different densities of particles. Denser particles, illustrated in Figure 14, represented a volume formed by the intersection of all mushrooms. As most of the mushrooms intersected within the highlighted area, particles densely populate this region. Consequently, the probability of a new mushroom growing in this area was very high. Conversely, areas with fewer mushroom intersections had less densely packed particles, indicating a lower probability of new mushroom growth in those areas. Lastly, particles were generated outside the mushroom geometries, representing an area of the lowest probability. This region, which accounted for the variability in the form of newly grown mushrooms—potentially exceeding the size of those already present, was considered. To illustrate this, particles were allowed to pop out from the mushroom area (or the initial balloon), but with restrictions on how far they could extend. The degree of extension was controlled by defining their stickiness in Maya, creating a visual representation of the larger mushroom population.

Although the probability space models in Figures 15 and 16 are analytical diagrams rather than predictive ones, they represent an exploration of a 3D method. The previous 2D drawings of expectations (Figures 3, 4, and 5) could be extrapolated further by taking the principles to construct three-dimensional representation models. This, in turn, might lead to the development of a 3D model utilising a point cloud system to forecast the probability space for a newly growing organism. For further investigation and the construction of a 3D predictive model, the following steps could be considered (Figure 17):

1. Use a volumetric matrix at a particular resolution as the base for the model.
2. Question the probability of a specific pixel containing an organism (such as a mushroom) during its growth process.
3. Generate a map, such as a 3D heat map, illustrating the ends or edges of the organism. This map would demonstrate the probability of each pixel containing the organism, with some pixels being more likely hosts (red) while others are less likely (blue) or not at all likely to contain the organism.

However, it is important to note that building such a model would require substantial data and computational resources, as well as sophisticated algorithms for processing and analysing the data. Therefore, this model will not be delivered in this PhD research due to the limitation of this study.

4.2. The New Role of The Designer and Living Material

Mushrooms, in specific morphologies, served as targeted design outputs. As the designer,

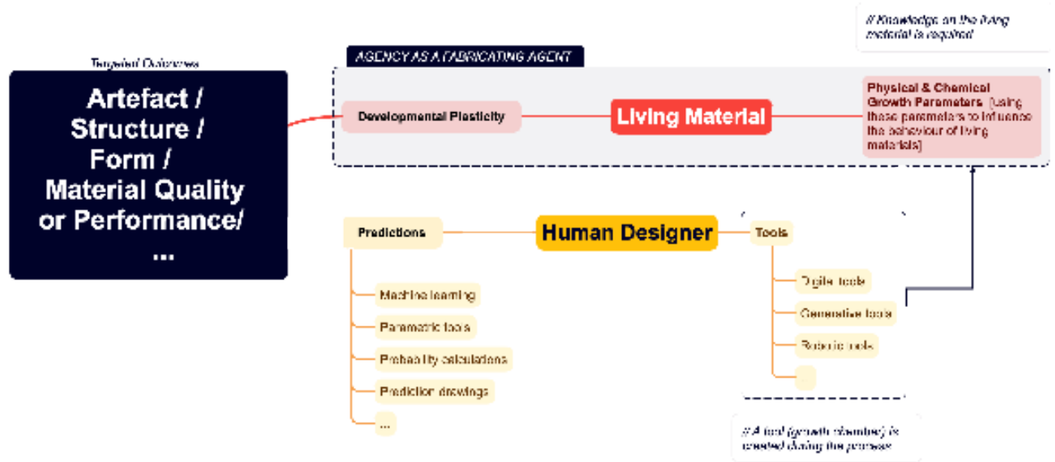


Figure 18

The diagram of the biofabrication process defined by harnessing the agency of living materials to create artefacts.

my goal was to activate the developmental plasticity that led to these morphological changes, driven by the mushrooms' inherent adaptability. My initial role involved configuring the growth conditions within a growth chamber to encourage the development of desired morphologies. Anticipating outcomes based on prior experience with the living material allowed me to guide its growth in a desired direction, within the limits of its developmental plasticity. This process required a deep understanding of the factors influencing these living materials and their developmental pathways. Such factors became critical parameters for designers to engage with when creating new structures or artefacts.

While the mushrooms grew, they actively altered the preset conditions through their interactions with their environment. Their role extended beyond passively responding to inputs; they dynamically influenced the growth conditions during their biological developmental processes. This dynamic created a cybernetic system, where the participants -designer, tools and living material- mutually influenced one another, transforming the process into an interactive, process-driven collaboration with living systems (Pask 1961). This collaboration involved distributed decision-making, adaptability, and dynamic interactions between the design output and their environments, operating without centralised control.

As a result, the designer's role in form-making shifts when working with living materials. The predominance of a predefined shape over the material is not feasible, particularly in bottom-up fabrication methods while designing with living systems. Unlike traditional design, where designers have significant control over the final shape of an object, in biodesign, the material and designer share a more balanced relationship. Here, the designer collaborates with the material rather than exerting dominance, embracing the agency of the living system (Figure 18).

This decentralised control, as demonstrated by the design experiments, revealed that designers' intentions and material outcomes do not consistently align, primarily due to the agency of living materials. The nonlinearity of these materials poses a significant challenge for designers incorporating them. Consequently, the design experiments helped enhance designers' comprehension of how the material behaves across diverse environmental conditions, aiming to reduce uncertainty in their designs.

Finally, when designing with living materials, the design target in such processes is multi-faceted. While one primary outcome is the cultivation of a living material with specific functional or morphological properties and characteristics, the process equally emphasises the development of a broader framework for interacting with living systems. Consequently, the design outputs extend beyond the material artefact (e.g. mushrooms) and included: (i) the bespoke tools built for specific purposes, such as growth chambers designed to regulate the growth environment, (ii) probability drawings or diagrams that mapped potential outcomes, and (iii) a functional cybernetic system, along with a design framework and methodology that themselves constituted a design output.

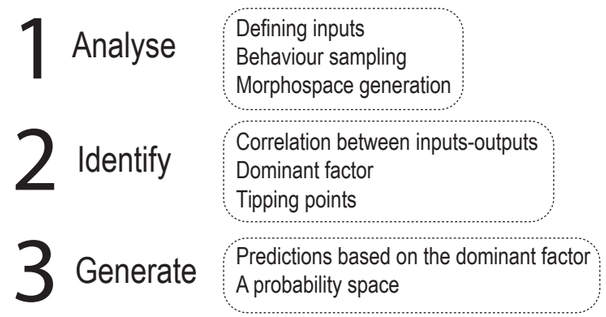
5. Conclusion

This explored the feasibility of establishing a design process that enables designers to predict the form of their material under untested conditions. To address this issue, design experiments were conducted to discover the potential of utilising the input-output relationship for predictive purposes. The results demonstrated that the morphologies of mushrooms exhibit a certain degree of predictability, based on their nonlinear correlational morphological behaviour across various environmental inputs. After understanding their level of predictability, the chapter proposed a 'probability space' method applicable in the design process involving living materials. This method acknowledged a significant margin of error, given that the morphology of living materials emerges as a result of complex processes.

The proposed probability space method, presented by developmental diagrams, aimed to identify zones including both low and high growth likelihoods, considering diverse growth possibilities for mushrooms. To generate the probability space for a form-giving organism, the following steps became essential: (i) analysing the environmental input and morphological output relationship; (ii) identifying the type of correlation between them, considering dominant factors and tipping points; and (iii) generating predictions based on the outcomes of the previous two steps (Figure 19). By using a method to predict the development of living materials, designers can make more informed decisions about how to manipulate the inputs to achieve desired outcomes. For example, if the anticipated results from the drawings or models do not align with the designer's expectations, adjustments to the experiment parameters can be made in advance. As a result, the form of the studied organism will closely align with the intended design.

A change in the designer's decision-making process, moving away from relying only on intuition and other biases and instead incorporating an automated method, can lead to the creation of BioCAD software. This BioCAD system could have the capacity to make predictions based on observed material behaviours and might be used as a design method and tool. As suggested by Gill Sunshine in his thesis, the development of new modelling tools is essential to address material irregularities and respond to emerging material realities (Sunshine 2022).

Figure 19
A list of steps that could be followed to demonstrate how to design a living material based on predictions.



Finally, designing based on the material probabilities, even with inaccuracies, results in a shift in the designer's role within the design process. As Carpo points out, although co-designing with nature was once considered a romantic thought from the 19th century, the dream becomes achievable in the 21st century (Carpo 2013), thereby altering the designer's role in the overall process. The designer now focuses on designing within the probabilities rather than certainties, transforming from a traditional, control-oriented creator to a facilitator and collaborator. They navigate within the limitations of the materials, acknowledging and embracing a wide range of errors and probabilities during the process.

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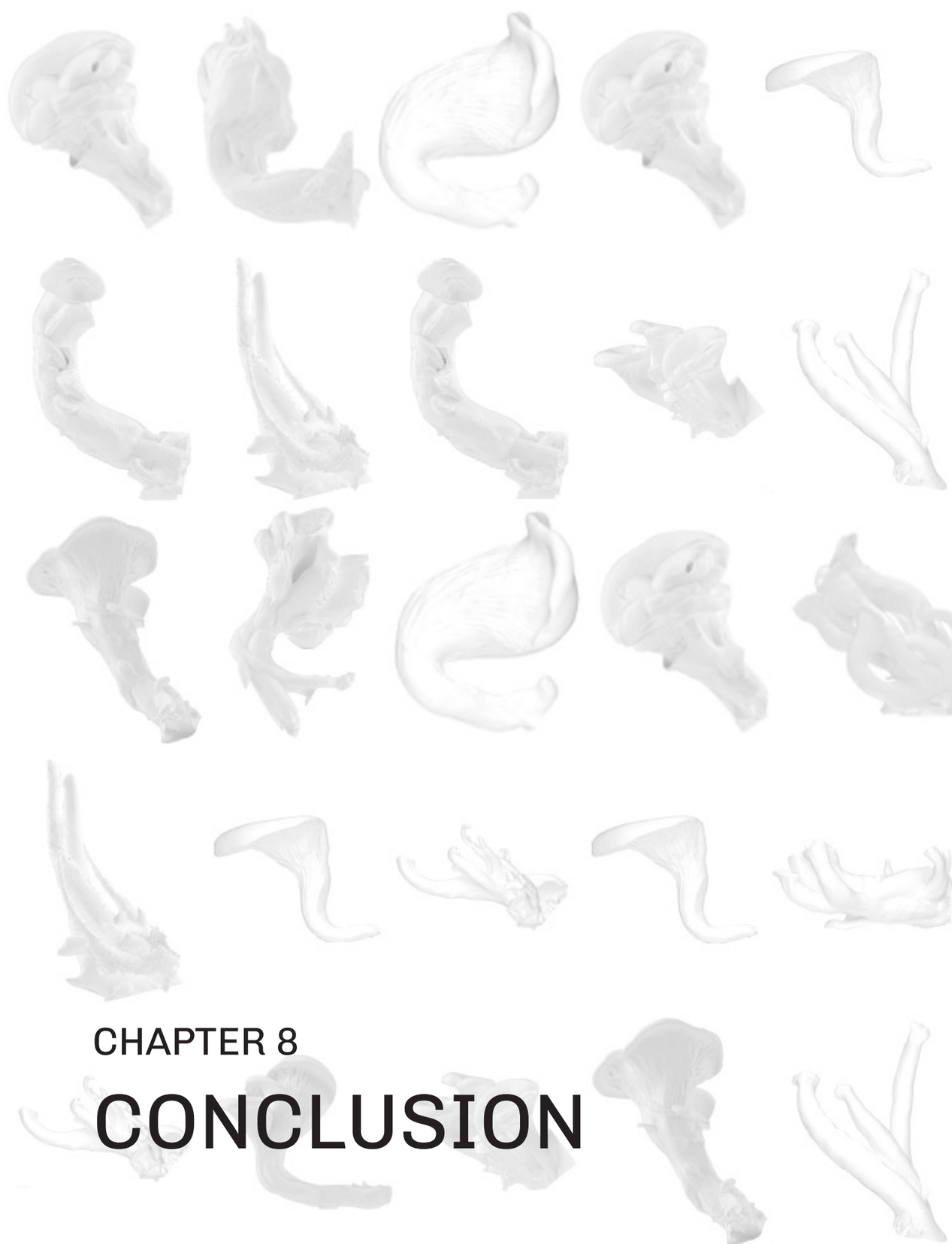

CHAPTER 8

CONCLUSION

1. A Framework for Parametric Biological Fabrication

This research aimed to develop a design framework and biodigital fabrication method to guide the growth of a living material that facilitates interaction between the designer, the material, and the tools used in digital architectural practice. To achieve this goal, the research started with an examination and redefinition of the relationship between a living material and design practices within the realm of biodesign. Key concepts such as New Materialism, form generation, top-down and bottom-up fabrication approaches and material emergence were investigated. Subsequently, the notion of “making-through-growing” emerged as a central concept for fabricating living materials (Table 1).

To develop a biofabrication tool and method that is applicable to making-through-growing, the study proposed a parametric system for biological fabrication. Understanding the processes of digital and biological form generation, as well as the direct and indirect associations between input parameters and output form helped to develop a “biological parametricism” approach. The significance of the proposed parametric biofabrication method lied in its transformative impact on the design process. It used the metabolism of organisms to design and fabricate living artefacts. This approach made a shift in the way designers create, and aligns with New Materialist theories that allow materials to perform their tendencies and capacities.

The proposed method was achieved through a series of form-making exercises using indirect morphological influences. Minimal physical intervention was achieved through a digitally controlled growth chamber, which allowed the organism to give form.

The process then involved the understanding of key points creating the framework, including:

i. Control points: Control points are factors that have an indirect (nonlinear correlational) influence on the morphology. These factors are determined based on the nature of the organism and then become input parameters for designers to interact with for fabrication purposes.

ii. Related control points: It was then realised that the influencing factors are not independent; rather, they are related to one another. In systems with interconnected control points, modifying one variable affects others, as well. These interconnected relations are the main factor complicating parametric biological behaviours.

iii. Cyber biological system: The control points related to influencing the growth possibilities of the organism are informed by a digital system. The organism, in turn, reflects back to this digital system by changing the parameters and influencing its environment; meaning that a feedback loop transitions into a cybernetic process when the organism begins to both inform and regulate its environment. The simple digital tool turns into a sophisticated (biological cybernetic) system, with a living organism or a biological brain.

iv. A parametric biological system: The combination of multiple interconnected elements, including indirect factors, developmental plasticities, interrelated parameters, and cybernetics through the organism's response constitutes a parametric biological system.

v. Boundaries of growth possibility: Although control points are interconnected, the interrelated relationships between form and parameters can be predicted within certain probabilities. These probabilities, defining the morphological features, are set within boundaries that cannot be crossed due to the natural limits of the organism or conditions that trigger a shift in the organism's behaviour.

This holistic and dynamic framework emphasised the organism's ability to respond and adapt to designed factors, highlighting the complexity and interdependence within biological systems. As a result, predicting the effect of altering multiple variables on an organism's morphology, and understanding and extrapolating the proportional relationships among these variables, might be an answer to the question of how designers can craft biology. Understanding how to influence the form of organisms also embodies a physical manifestation of a 'Creodic' design process, searching for the necessary pathways for living cells to create materials towards a direction that aligns with desired outcomes (Dade-Robertson 2021).

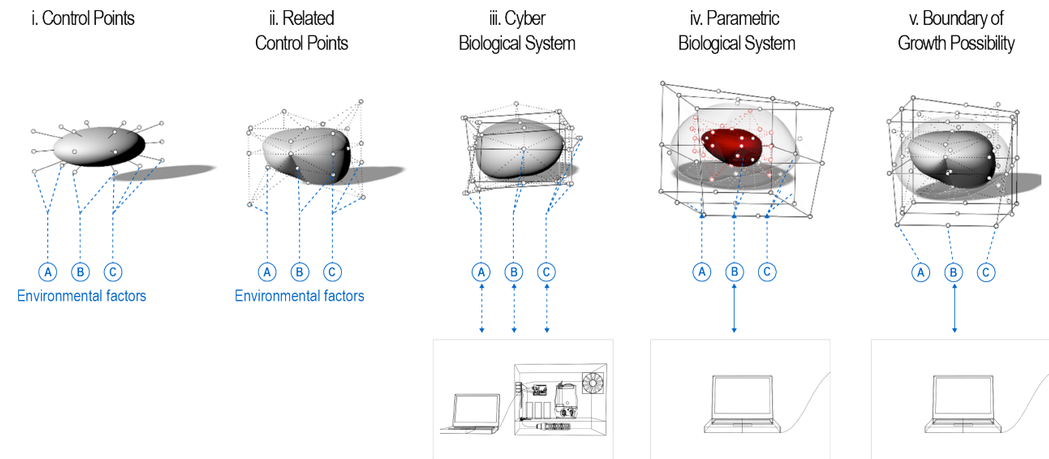

Figure 1

Diagram illustrating the elements that create the framework of a parametric biological system.

Table 1

Mapping research objectives with corresponding findings.

Objectives	Findings	Chapters
To examine, understand, and redefine the relationship between living materials and design practices in the context of biodesign.	The notion of making-through-growing emerged as a central concept for fabricating living materials.	C3: New Materialism
To develop a biofabrication tool and method that: (1) is applicable to “making-through-growing”,	The direct and indirect relationships between input parameters and output form was incorporated into biofabrication as a design principle.	C4: Parametricism
(2) allows material interaction through its developmental parameters, (3) allows designers to work with the tendencies and capacities of living materials.	The developmental plasticity of organisms was employed as a fabrication method, with the chemical environment used to guide the growth of mycelium. Through design experiments, various qualities of fungi, including their form-taking and form-giving abilities at micro and macro scale, were discovered.	C5: Plasticity and Form Taking
	A digitally controlled growth chamber was built and used as a parametric tool for fabricating fungal fruiting bodies within the physical environment. A nonlinear correlational relationship was observed among tipping points of the organism, helping in the development of a parametric biological fabrication method. It was understood that while each environmental factor influences the overall morphology, certain factors exhibit a stronger correlation with the output form, thus being identified as dominant factors.	C6: Linearity and Form Giving
To develop a design concept (based on predictions), presenting the methods for designing living materials under untested conditions.	Dominant factors were used to predict the shape of a mushroom for conditions that haven’t been tested yet and a probability space method was proposed that represents the range of possible outcomes for a given set of conditions.	C7: The Probability Space

The creativity in this research lay in its reframing of design paradigms and its systems-oriented approach that allows for biological agency. By applying the computational design concept of “parametrics” to a non-linear, living material, the work challenged traditional approaches to material design. It achieved this by integrating the living material, the designer, and the environment into a dynamic feedback loop system. Building such a system without centralised control required weaving together diverse interdisciplinary components, resulting in a novel experimental framework. This was achieved through iterative experiments, where fundamental concepts such as tolerance, tipping points, thresholds, linearity, and probability are rediscovered and recontextualised within this new paradigm. The originality of the research lay in its ability to apply these established ideas to an entirely new domain, pushing boundaries, fostering innovation, and prompting a rethinking of traditional design and material frameworks.

2. Reflections on Methodology

Implementing the mixed methods methodology posed several challenges in this research, especially when applied within a multidisciplinary framework that integrated the scientific method. The research involved collaboration across diverse fields, such as architectural design and science/mycology. This interdisciplinary nature introduced complexities in uniformly applying a single scientific method throughout the design process. For instance, when conducting experiments with fungi, the need for a rigorous setup with a specific recording/measuring method posed challenges. The experiments required the management of numerous interconnected variables to accurately isolate the impact of specific factors. However, this was challenging due to the inherent complexity of living materials and their responses to various conditions. These challenges made adhering to the traditional scientific method’s high level of control difficult in experiments involving living systems. To ensure rigour, a growth chamber was constructed for better control over variables. However, in certain instances, the designer still had to intervene manually within the experimental system to maintain controlled conditions. Ultimately, while adapting scientific methods for design presented challenges, these obstacles led to the development of bespoke methods and approaches for interdisciplinary contexts.

The design research often proceeded non-linearly, making it difficult to set up all design experiments in advance. Predicting outcomes or determining cause-and-effect relationships between design variables and outcomes was challenging due to the involvement of multiple interacting variables and factors when designing with organisms. In the research through design process, the output of one experiment often sparked new questions and insights, and the next step was not always clear. This necessitated a flexible approach to design research, where I was open to exploring new avenues and adjusting my plans based on the results of their experiments. Each chapter presented new questions stemming from the findings of the previous one. Despite these challenges, design researchers may explore the complexities of the design process and gain a deeper insight into the factors influencing design outcomes.

3. Current Limitations

3.1. Applicability

The current scope of the research, focusing on the use of developmental plasticity in organisms for their fabrication, is currently limited to a specific subset of biology. However, there is potential for its extension across a broader range of species. This approach, which involves minimal physical interaction, is primarily applicable to form-giving organisms, such as mushrooms, plants, or organisms capable of constructing macro structures, although it may also apply to form-taking organisms such as mycelium, algae, or bacteria. However, form-taking organisms may necessitate an external structure or mould for attachment and growth at a macro scale.

Parametric behaviour is associated with nonlinear correlations, and although it enables the designer to predict material behaviour, there is a significant margin of error that could impact design decisions.

Lastly, once the growth fabrication process is completed, there is a need to care for the biological material to maintain its form. This limitation is related to working with living systems in general, rather than being specific to the proposed bio-digital fabrication method. It becomes a critical consideration for the designer during the process to make decisions on how to sustain the organism's current state and make that state/form/performance long-lasting for potential applications.

3.2. Accuracy

The accuracy of the proposed probability space models is limited by the amount and quality of data available to inform those models. However, increasing the number of samples can help to improve the accuracy of the models by providing more data for analysis. Additionally, the interpretation of probabilistic results may be challenging for designers who are not familiar with probability theory. It is important for designers to work closely with data analysis and probability to ensure that the results are properly understood and used in the design process.

Lastly, the use of analytical diagrams to investigate 3D techniques is limited and not automated. Although these diagrams help develop 3D models that represent the probability space for a growing organism, there is a need to further extrapolate from 2D forecasts. A 3D model that predicts the probability space for a growing organism based on a point cloud system, could help create a more accurate representation of the organism's growth.

4. Contrasting Digital and Biological Form Generation

Within this thesis, the investigation of using digital form-generation techniques for designing biological artefacts has shed light on the nuanced differences between digital and biological form-generation domains, particularly regarding responsiveness, reliability, repeatability, and controllability (Table 2). In the digital domain, objects such as NURBS (non-uniform rational basis spline), blobs or hyper-surfaces are passive and depend strictly on the designer's commands. On the contrary, biological objects can actively respond to external forces and be involved in the formation process. However, they do not exhibit immediate visible responses to changing parameters. Instead, they require a growth time before any effects of altered parameters become apparent. Thus, time becomes a more prominent factor when working with biological materials. More importantly, the level of computation or calculation differs significantly in digital and biological systems. Digital models only operate within predefined calculations and do not account for unexpected consequences. In these models, every potential outcome arises from the decisions made by the designer and the software.

Table 2

A summary of the similarities and differences between working with a digital object and a living cell.

	Making Digital Form	**Making-through-Growing Biological Form**
Fabrication Medium	Software	Bioreactors, growth chambers, synthetic biology
Design Space	Topological space	Phase space and physical space
Morphological Modifications	Through parameters and algorithms, formed by the designer	Through parameters formed by its agency and the designer's intervention in physical, genetic, or environmental factors
Variables	Only defined variable	Everything can be a variable, too sensitive
Responsiveness	Cannot be cybernetic	Can be cybernetic and an organism can influence the fabrication process
Reliability	Linear or Nonlinear. It can be set by the designer (by including random factors)	Nonlinear with a correlational or non-correlational

In contrast, biological materials are subject to multifaceted factors extending beyond designer definitions, often intricately linked. The smallest details can influence the organism, and due to the inherent complexity and intelligence of living systems, outcomes become more diverse. Alterations in one variable may unpredictably affect others, and stabilising a variable constant may be challenging due to living systems' inherent tendency to achieve equilibrium. Moreover, within living systems, varying a parameter may not consistently produce the same outcome. While the parametric approach helps to manage complex systems, the reliability and repeatability of biological materials make them challenging to work with when attempting to apply a purely parametric approach with direct associations.

5. New Roles and Relations of Designer, Material and Digital Tools

The contribution of this thesis in the development of a parametric system for biological fabrication has also changed the traditional roles of a designer, material, and digital tools. Con-

sidering the aforementioned factors, although the design process of biological and digital objects exhibits commonalities, particularly in the application of parametric design methods, the complexity of working with living systems introduces additional layers. Unlike designing with digital objects, which are more within the control of designers, the process is not straightforward when working with living systems. Adding minimal physical control in the process, allowing for material emergence, causes a change in the role of a designer from a control-oriented creator to that of a facilitator.

Designers, in such processes, work within the limitations of the materials, accepting that complete control is not possible. Rather than trying to dominate every aspect of the process, designers adapt to working with irregularities, iterations, unexpected material behaviours, nonlinearities, probabilities, and predictions. Therefore, a designer tries to understand the behaviour of materials which are intricately tied to various aspects of their environment, forming different types of input-output relations. The designer works as a gardener, botanist, adjuster, bio-craftsperson, guider, or predicter focusing on interdependent components and processes that work together to perform a particular function. The verbs -- nurturing, guiding, growing, crafting, or sculpting -- capture different aspects of this intricate process.

In biodesign processes, where living materials are not obedient, a material's role undergoes a significant change, impacting the design processes. Since not all living materials can receive form through their developmental plasticity, designers face limitations in shaping these materials according to their morphological limitations. However, when designers employ fabrication methods that enable the emergence of form, these living materials become co-designers in the process. This means that instead of designers rigidly defining their form, these materials have the ability to evolve, thereby contributing to the shape of the final design.

Other than the change in the designer's and the material's role in parametric biofabrication processes, the role of digital tools also transforms. Fabricating living materials using digital tools bridges the gap between coding languages and biological forms. The interaction of biological and digital systems has the potential to initiate new cybernetic interactions. Within this symbiotic relationship, living organisms emerge as active participants in the fabrication process, acting as non-human controllers that influence the design setting. It is important to note that cybernetic digital systems should not be seen as a replacement for the designer's expertise and intuition. Despite the complex biohybrid relations, ultimately, the designer is responsible for making micro decisions that shape the final product.

6. Recommendations for Future Work

6.1. Machine Learning

Future research plans may focus on understanding and predicting the behaviour of materials with nonlinearities for practical real-world applications. This exploration could be done on two levels: (i) on a micro scale, particularly for living building materials; and (ii) biocomposites capable of self-assembly, responding over time through processes such as shrinking, growing, or changing its developmental state. Working with such complex or unfamiliar systems may be challenging for a designer. Therefore, predicting their response in advance and understanding the key factors to control/guide their behaviour in real-world applications becomes necessary.

Managing the uncertainty associated with these materials necessitates the creation of computational models. These models can help in the analysis and understanding of the underlying mechanisms and processes governing their behaviour. In particular, the research agenda should not be limited to building more accurate and controllable prediction models for an individual material. Instead, there is a need to develop a comprehensive system with digital twins and a library documenting the behaviour of various materials.

One strategy to achieve successful material behaviour predictions, to some extent, involves the use of an additive system, such as Machine Learning or Artificial Intelligence. For example, a combination of image-based, marker-based, and pin-based methods allows designers to collect data and enhances the intelligence of their digital models (Rossi et al. 2021). An alternative method for data generation and collection is the use of a real-time sensory feedback system equipped with various sensors (Im et al. 2018). After understanding the key factors in controlling the material behaviour, training Machine Learning models can be used to generate predictive models. There are numerous examples of AI-based predictive models; however, their application in the field of design is not yet widespread.

6.2. CAD for Living Materials

The next step of predictive modelling that will allow designers to make more informed decisions about how to manipulate the inputs to achieve desired outcomes may be software development. Current CAD software used for creating interactive 3D animations, models, and simulations is based on traditional, standardised, and linear material production. A notable limitation lies in the fact that their simulation capacities are not sufficiently advanced for irregular, nonlinear, adaptive, or responsive material systems. Therefore, there is a need for a geometric paradigm necessary to respond to emerging material realities (Sunshine 2022).

The development of 3D modelling software tailored for architecture, specifically designed to handle under-processed and irregular materials, holds the potential to streamline the manual calculations associated with material behaviour. One such innovative solution is the BioCAD software, anticipated for engineering simulation and analysis. This software might have the capability not only to understand and optimise material systems, but also feature visualisation and rendering capabilities. This alternative contemporary CAD software that effectively represents the potential of materials would empower designers to make well-informed decisions during the design process, enabling them to manipulate inputs strategically to achieve desired outcomes.

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APPENDIX

1. Biological Control of Substances Hazardous to Health (BioCOSHH) form

Ref:

Newcastle University
Microbiological Hazards and Genetic Modification Safety Advisory Sub-Committee

BioCOSHH Risk Assessment

A BioCOSHH risk assessment is required for work with biological agents and hazards. The form should be completed electronically and signed by the principal investigator. Hazard Group 2 and 3 biological agents and hazards must be registered using the Pathogen Registration form and the BioCOSHH form sent by email to the University Biological Safety Officer. The possession or use of any Hazard Group 3 biological agent or the Hazard Group 2 biological agents *Bordetella pertussis*, *Corynebacterium diphtheriae* and *Neisseria meningitidis* requires permission from the University Biological Safety Officer. Guidance on completing this form is provided in the BioCOSHH Risk Assessment section of the Safety Office website.

Title of project	Guiding the growth of mycelium
Principal investigator / Responsible person	Prof Martyn Dade-Robertson / Dilan Ozkan
School	School of Architecture, Planning and Landscape, HBBE
Date of assessment	05.02.2019
Location of work (Buildings and room numbers)	Devonshire Building 5.7

Section 1 Project or Activity

1.1: Brief description of project or activity

Growing mycelium on tow various substrates (i. (PDA) agar and ii. Medium (including white vinegar, sodium bicarbonate, salt) caused shrinkage on mycelium growth) and investigating the mycelium growth and interactions to develop a new biological fabrication strategy.

Pleurotus Ostreatus spawn ordered from Urban Farm-It, UK

Section 2 Hazards

2.1: Biological agents or hazards

Pathogens (ACDP/DEFRA Hazard Group 1)	*Pleurotus ostreatus* *Most species of Pleurotus are plant pathogenic, causing root and stem rot on a variety of monocots, dicots, and gymnosperms, which results in the death of affected trees. Some species are saprophytic and cause white rot of wood.*
Pathogens (ACDP/DEFRA Hazard Group 2)	[ENTER DETAILS HERE]
Pathogens (ACDP/DEFRA Hazard Group 3)	[ENTER DETAILS HERE]
Toxins	Ethanol, White vinegar, Sodium bicarbonate, Salt
Carcinogens	[ENTER DETAILS HERE]
Allergens	[ENTER DETAILS HERE]
Human primary or continuous cell cultures	[ENTER DETAILS HERE]
Animal primary or continuous cell cultures	[ENTER DETAILS HERE]
Human cells or tissues	[ENTER DETAILS HERE]
Animal cells or tissues	[ENTER DETAILS HERE]
Human blood	[ENTER DETAILS HERE]
Patient contact	[ENTER DETAILS HERE]
Animals	[ENTER DETAILS HERE]
Plants	[ENTER DETAILS HERE]
Soils	[ENTER DETAILS HERE]
Other biological hazards	[ENTER DETAILS HERE]
[ENTER DETAILS HERE]	

Section 3 Risks

3.1: Human diseases, illnesses or conditions associated with biological agents or hazards

N/A. The fungi are class 1.

3.2: Potential routes of exposure

Inhalation ■ Ingestion ■ Injection ☐ Absorption ☐ Other ☐ — Select all that apply

[ENTER DETAILS HERE]

3.3: Use of biological agents or hazards

Small scale ■ Medium scale ☐ Large scale ☐ Fieldwork ☐ Animals ☐ Plants ☐ Other ☐ — Select all that apply

Mycelium will be grown on a substrates like sawdust and hemp shives up to 10 kg
Mycelium will be grown on malt extract agar plates.

3.4: Frequency of use

Daily ☐ Week ■ Monthly ☐ Other ☐ — Select one

[ENTER DETAILS HERE]

3.5: Maximum amount or concentration used

Negligible ☐ Low ■ Medium ☐ High ☐ — Select one

[ENTER DETAILS HERE]

3.6: Levels of infectious aerosols

Negligible ■ Low ☐ Medium ☐ High ☐ — Select one

The fungi are class 1and not pathogenic.

3.7: Potential for exposure to biological agents or hazards

Negligible ☐ Low ■ Medium ☐ High ☐ — Select one

All experimentations will be performed in the lab environment.
While both the fungi are class 1, other organisms can grow on the substrate, if exposed to environment.
Mycelium can grow on wooden structures if in close contact.

3.8: Who might be at risk (*Contact the University Occupational Health Service)

Staff ■ Students ■ Visitors ■ Public ☐ Young people (<18yrs) ■ *New and expectant mothers ☐ Other ☐

The main risks will be to those doing the work and to other users of the lab and storage facilities.

3.9: Assessment of risk to human health (Prior to use of controls)

Level of risk — **Effectively zero** ■ **Low** ☐ **Medium/low** ☐ **Medium** ☐ **High** ☐ — Select one

3.10: Assessment of risk to environment (Prior to use of controls)

Level of risk — **Effectively zero** ☐ **Low** ■ **Medium/low** ☐ **Medium** ☐ **High** ☐ — Select one

Section 4 Controls to Reduce Risks as Low as Possible

4.1: Containment

Laboratory ■ Animal facility ☐ Plant facility ☐ Other ☐ — Select all that apply

[ENTER DETAILS HERE]

4.2: Containment level

Containment level (CL 1) ■ Containment level (CL 2) ☐ Containment level (CL 3) ☐ — Select one

All experiments will be performed in the lab environment.
While both the fungi are class 1, other organisms can grow on the substrate, if exposed to environment.
Mycelium can grow on wooden structures if in close contact.

4.3: Microbiological safety cabinets (MSC)

Class 1 ■ Class 2 ☐ Class 3 ☐ Other ☐ — Select all that apply

[ENTER DETAILS HERE]

4.4: Other controls

PPE is worn at all time and the use of sharps will be avoided unless there is no suitable alternative in which case safe work practices will be used to prevent sharps injuries which might lead to exposure to pathogens. Hands will be washed after the work activity is completed.

4.5: Storage of biological agents or hazards

In the fridge or incubator.
Mycelium spawn will be stored in the fridge up to 3 months.
White vinegar, salt and sodium bicarbonate will not be stored in the lab.

4.6: Transport of biological agents or hazards

Samples are bagged for transport or kept in closed containers.

4.7: Inactivation of biological agents or hazards

Disinfection ■ Autoclave ■ Fumigation ☐ Incineration ☐ Other ☐

Disinfection and autoclaving will be carried out where required. All waste materials will be disposed of into the autoclave bags, yellow clinical waste bags or sharps bins as required. All used sharps will be placed immediately after use into a sharps bin. Sharps bins will be located on the bench where the sharps are used so that they can be disposed of directly after use. All waste will be disinfected before disposal.

a) Disinfection.
70% Ethanol will be used for disinfection. The surfaces of the microbiological safety cabinet will be disinfected with 70% Ethanol before and after use. Laboratory benches will be swabbed with 70% Ethanol after any activity.

1% Virkon will be used for disinfection. The surfaces of the microbiological safety cabinet will be disinfected with 1% Virkon after use. Laboratory benches will be swabbed with 1% Virkon after any activity.

b) Autoclaving.
All contaminated materials, including waste destined for incineration, will be inactivated by autoclaving (100% kill) prior to disposal of waste or cleaning and recycling of reusable laboratory equipment, such as glassware.

4.8: Personal protective equipment (PPE)

Lab coat ■ Lab gown ☐ Surgical scrubs ☐ Disposable clothing ☐ Apron ☐ Spectacles ☐ Goggles ☐ Face shield ☐ Gloves ■ Special headwear ☐ Special footwear ☐ Other ☐	Select all that apply

Lab coats and disposable nitrile gloves will be worn to carry out this work.

4.9: Respiratory protective equipment (RPE)

Disposable mask ☐ Filter mask ☐ Half face respirator ☐ Full face respirator ☐ Powered respirator ☐ Breathing apparatus ☐ Other ☐	Select all that apply

Not required

4.10: Health surveillance or immunisation (If you need advice contact the University Occupational Health Service)

Not required.

4.11: Instruction, training and supervision

All workers will be properly trained and monitored by the principal investigator and suitably qualified laboratory supervisors and no worker will be allowed to work unsupervised in the laboratory until they are suitably and sufficiently trained. Training procedures, which include information on hazards and risks and control measures to be used. Copies of all risk assessments and standard operating procedures will be kept in the laboratory and reviewed regularly and immediately if there are any changes to the risks or the nature of the work.

All workers will be appropriately trained in procedures and safe work practices. All workers will be appropriately supervised until they are competent to safely perform unsupervised all aspects of the work including the emergency procedures. Access to the lab is restricted to personnel approved by the principal investigator.

The principal investigator will carefully monitor all activities to ensure that all control measures are properly and fully implemented and all workers are suitably trained and competent to safely perform the work in the laboratory.

4.12: HSE consent or DEFRA licence

N/A

Section 5 Emergency Procedures

5.1: Emergency procedures

Spillages of any *spawn* or any of their contaminated products will be autoclaved and disinfected with 1% Virkon or 70% ethanol.
In the event of any injection injury or other serious accidental exposure seek first aid or medical attention if required.

5.2: Emergency contacts

Name	Position	Telephone
Dilan Ozkan	PhD Student	
Prof Martyn Dade-Robertson	Principal Investigator	

Section 6 Approval

6.1: Assessor

Name	Signature	Date
Dilan Ozkan	Dilan Ozkan	05.02.2019

6.2: Principal investigator / Responsible person

Name	Signature	Date
Prof Martyn dade-Robertson		

Risk Estimation Matrix

Severity of harm	Likelihood of harm			
	High	**Medium**	**Low**	**Negligible**
Severe	High	High	Medium	Effectively zero
Moderate	High	Medium	Medium/low	Effectively zero
Minor	Medium/low	Low	Low	Effectively zero
Negligible	Effectively zero	Effectively zero	Effectively zero	Effectively zero

The University will process the data provided on this form in accordance with the HR privacy notice which can be found on our website at https://newcastle.sharepoint.com/hub/hr/news/Pages/GDPR-HR-Privacy-Notice.aspx

2. Control of Substances Hazardous to Health (COSHH) Risk Assessment Form

COSHH Risk Assessment

Newcastle University OHSS: H&S Form 401.1a

This form should be completed electronically and signed by the Principal Investigator or responsible person. Guidance on completing this form is provided in the COSHH Risk Assessment section of the OHSS website.

Section 1: Project Details

1.1.	**Title of project or activity**	Guiding the growth of mycelium		
1.2.	**Principal investigator/responsible person**	Prof Martyn Dade-Robertson / Dilan Ozkan		
1.3.	**School/Institute/Service**	School of Architecture, Planning & Landscape, HBBE		
1.4.	**Location of work** building and room numbers	Devonshire 5.7		
1.5.	**Brief description of work activity**	Growing mycelium on various substrates and investigating the mycelium growth to develop a new biological fabrication strategy. *Pleurotus Ostreatus* Spawn ordered from Urban Farm-It, UK		
1.6.	**Date of assessment**	05/02/2019	**1.7. Revision date***	

Section 2: Emergency Quick Reference

The purpose of this section is to provide easy access to emergency information. A full assessment of risk will be provided in the next sections and **completing this section last is advisable.**

2.1. Emergency contacts One of these should be the PI/responsible person Security can be contacted on extension 6666	**Name:**	Ben Bridgens	Dilan Ozkan
	Position:	PI	PhD Student
	Telephone number:		077798691165

2.2. Hazard pictograms – select all that apply to the work activity.

 *Health hazard*	 *Toxic*	 *Corrosive*	 *Harmful/ Irritant*	 *Flammable*	 *Oxidising*	 *Explosive*	 *Compressed gas*	 *Danger for the environment*
x		x	x	x	x			

2.3. Name of hazard	**2.4. Properties of hazard** Briefly describe how the chemical is hazardous e.g. toxic, flammable, carcinogen	**2.5. Emergency procedures** Include, as appropriate, procedures for: • Contained Spill • Small uncontained spill, • Large uncontained spill • First aid • Fire
Ethanol  	Highly flammable liquid and vapor Causes serious eye irritation	**Contained Spill:** Soak up with absorbent material and dispose of as hazardous waste. Wear gloves, lab coat and safety glasses Keep in suitable, closed containers for disposal. **Small uncontained spill :** Remove sources of ignition. Soak up with absorbent material and dispose of as hazardous waste. Wear gloves, lab coat and safety glasses Keep in suitable, closed containers for disposal. **Large uncontained spill:** Remove sources of ignition. Contain spillage, and then collect with an absorbent material and dispose of as hazardous waste. Wear gloves, lab coat and safety glasses Keep in suitable, closed containers for disposal. **First aid: If in eyes** rinse cautiously with water for at least 15 minutes. Remove contact lenses, if present and easy to do. Continue rinsing. **In case of skin contact** Take off immediately all contaminated clothing. Wash off with plenty of water. **If inhaled** move person into fresh air. Consult doctor if feeling unwell. **Fire:** Use water spray, alcohol-resistant foam, dry chemical or carbon dioxide. Water mist may be used to cool closed containers. **Special hazards arising from the substance or mixture.** Carbon monoxide (CO), Carbon dioxide (CO2).
Virkon   	Causes skin irritation. Causes serious eye damage. May cause respiratory irritation. Toxic to aquatic life with long lasting effects.	• **Contained Spill:** N/A • **Small uncontained Spill:** Same as above. If cleaning up small powder spill. Take up dry. Dispose of properly. Clean up affected area. Avoid generation of dusts. Keep in suitable, closed containers for disposal. • **First aid:** If in eyes rinse cautiously with water for at least 15 minutes. Remove contact lenses, if present and easy to do. Continue rinsing. In case of skin contact, take off immediately all contaminated clothing. Wash off with plenty of water. If inhaled move person into fresh air. Consult doctor if feeling unwell. • **Fire:** Use water spray, alcohol-resistant foam or carbon dioxide. **Special hazards arising from the substance or mixture.** Carbon oxides, Nitrogen oxides (Nox), Sulfur oxides, Potassium oxides, Sodium oxides Mixture with combustible ingredients. Development of hazardous combustion gases or vapours possible in the event of fire.

Additional rows can be added to this table as required

Section 3: The Risk Assessment

Additional rows can be added to this table as required

3.1. Name of hazard including substances and by-products produced during or as a result of the activity.	**3.2. Properties of hazard** Provide details of how the substance could cause harm. Useful sources of information are the safety data sheet for the substance, Hazard (H) statements (give the whole phrase not just the code), and the workplace exposure limit.	**3.3. Physical form** e.g. powder, dust, granular, pellet, liquid, solution, gas.	**3.4. Quantity and concentration** (give units)	**3.5. Frequency of use** e.g. daily, weekly, monthly, one-off.	**3.6. Route of exposure** e.g. ingestion, inhalation, skin/eye contact, skin absorption, injection/sharps injury.
Ethanol	H225 - Highly flammable liquid and vapor H319 - Causes serious eye irritation **Workplace exposure limit 1000 ppm or 1920 mg/m3**	Liquid	Max working solution 500 mL at 70%	weekly	inhalation, skin/eye contact
Virkon	H315 - Causes skin irritation. H318 - Causes serious eye damage. H335 - May cause respiratory irritation. H411 - Toxic to aquatic life with long lasting effects.	powder & solution	Supplied in x L container Max working solution 1% per L	monthly	inhalation, skin/eye contact
Salt	H318 - Causes serious eye irritation.	powder	Supplied in x L container	weekly	eye contact
White vinegar	H318 - Causes serious eye irritation.	liquid	Max working solution 500 mL at 5%	weekly	
Sodium bicarbonate	H318 - Causes serious eye irritation.	powder	Supplied in x L container	weekly	eye contact

3.7 Carcinogens All carcinogens and users of carcinogens should be notified to OHSS using the following link
https://newcastle.sharepoint.com/hub/orghas/Pages/chemical_carcinogens.aspx

3.8. Dangerous Substances and Explosive Atmospheres (DSEAR)	**Yes**	**No**
Are you carrying out an activity/chemical reaction that is at risk of thermal runaway or explosion?		**x**
Will the activity involve handling or storage of pyrophoric or unstable substances such as peroxide?		**x**
Will flammable vapours, solid particles, fibrous particles etc. capable of forming an explosive atmosphere be present in the working atmosphere?		**x**
If the answer to any of the above questions is yes, you will need to complete a short 'add-on' DSEAR risk assessment		

3.9. Who might be at risk? (tick all that apply)	**Staff**	**Postgraduates**	**Undergraduates**	**New or expectant mothers** (Contact Occupational Health)	**Contractors**	**Public including visitors and children**
	x	**x**		x		

3.10. Assessment of inherent risk to human health prior to the use of controls (please use the risk assessment matrix at the end of this form)	**High**	**Medium**	**Medium/low**	**Low**
				x

Specify for each hazard identified in section 3. Precautionary (P) statements are a useful source of information.	
4.1. Physical or Engineering Controls. LEV, fume hood, glove box, total containment etc. **Specify at which point in the work activity they are to be used.**	**Ethanol** to be decanted into 500ml spray bottles under fume hood and diluted with deionised water. Stored in flammable cabinet **Virkon** to be mixed into liquid media at the sink with running water to dilute any traces that occur in the sink.
4.2. Administrative controls Training requirements, access control, signage.	Biological and Chemical Safety Training required to any work activity in the lab. The lab has card access. Stock solutions will be made when needed and at a volume that should last some time – this will reduce frequency of exposure to concentrated/neat substances. Bottles labelled correctly
4.3 Personal Protective Equipment. Respirators, safety specs, face mask, lab coat, gloves etc. **Specify which type and when they are to be worn.**	A lab coat and nitrile gloves will be worn for all parts of the experiment. Nitrile gloves are compatible with all materials used.
4.4. Storage requirements Include a description of how hazardous substances including flammable materials will be stored. Describe how incompatible materials will be segregated.	Store in a cool location. Provide ventilation for containers. Avoid storage near extreme heat, ignition sources or open flame. Store away from foodstuffs. Store away from oxidizing agents. Store in cool, dry conditions in well-sealed containers. Keep container tightly sealed. Store in secure flammable storage area away from sources of ignition. Protect from freezing and physical damage.
4.5. Transport of the hazardous substance Describe how you will transport substances between laboratories or different university sites.	Hazardous substances should not be transported outside the laboratory. If needed substances must be in a container that is tightly closed and using a trolley.
4.6. Disposal procedures Carefully consider the safest means of disposal and identify when waste should be disposed of by a chemical waste contractor	In case of any waste, will be labelled as flammable and disposed of by hazardous waste disposal at the university.

	Yes	**No**	**Describe the findings of exposure monitoring or health surveillance**
4.7. Is exposure monitoring required? For example if you suspect that exposure to a chemical exceeds the workplace exposure limit. Contact OHSS for further advice		**x**	
4.8. Is health surveillance required? See Occupational Health surveillance policy and programme. Contact Occupational Health for further advice		**x**	

4.9. Assessment of residual risk to human health after the application of controls (please use the risk assessment matrix at the end of this form)	**High**	**Medium**	**Medium/low**	**Low**
				x

Section 5: Approval

I confirm that this is a suitable and sufficient risk assessment for the above described work activity	**Name**	**Signature**	**Date**
Assessor This is the person who has completed this form	Dilan Ozkan	Dilan Ozkan	05/02/2019
Principal Investigator/responsible person	Prof Martyn Dade-Robertson		

Risk estimation matrix Use this to complete sections 2.10 and 3.10

Severity of Harm	**Likelihood of harm**		
	High	**Medium**	**Low**
Severe	High	High	Medium
Moderate	High	Medium	Medium/low
Minor	Medium/low	Low	Low

Please keep a record of this risk assessment

***Review of assessment**

This assessment should be reviewed every 2 years and immediately if there is reason to believe that it is no longer valid (e.g. after an accident/incident), if there is a significant change in the work activity to which it relates or if the results of monitoring or health surveillance indicate it to be necessary.

3. Arduino code

AllTogether

```
//  pinMode(trigPin, OUTPUT);//Distance sensor
 // pinMode(echoPin, INPUT);

  Serial.begin(100);
  dht.begin();

analogReference(DEFAULT);

}

void loop(){
   delay(500);
  float h = dht.readHumidity();
  float t = dht.readTemperature();

  if (isnan(h) || isnan(t)) {
    Serial.print("fail to read");
    return;
  }

  Serial.print("humidity is=");
  Serial.println(h);
  Serial.print("temperature is=");
  Serial.println(t);

  if (digitalRead(HUMIDIFIER) == ON)
  {
    if (h > SETPOINT + DEADBABD)
    {
      digitalWrite(HUMIDIFIER, OFF);
    }
  }

  else
  {
    if (h < SETPOINT - DEADBABD)
    {
      digitalWrite(HUMIDIFIER, ON);
    }
 }
    if (digitalRead(HEATBULB) == ON)
    {
      if (t > MAXTemp )
```

AllTogether

```
#include <DFRobot_DHT11.h>

//https://www.youtube.com/watch?v=DAn4UguyzfE
//https://www.youtube.com/watch?v=AEJ8MQyEOBg


#include <DHT.h>//Humidifier
#define DHTPIN 2
#define DHTTYPE DHT11
DHT dht(DHTPIN, DHTTYPE);
#define SETPOINT 95
#define DEADBABD 1
#define HUMIDIFIER 7 //where to pin
#define ON false
#define OFF true

#define MAXTemp 21 //Heat bulb
#define MINTemp 19
#define HEATBULB 8 //where to pin
#define ON false
#define OFF true

int sensorPin = A1;//Fan
int motorPin = 4;
int sensorVal;
int motorPinVal;

//const int trigPin=11;//Distance sensor
const int echoPin=10;
float duration, distance;


void setup(){
  pinMode(HUMIDIFIER, OUTPUT); //Humidifier
  digitalWrite(HUMIDIFIER, OFF);

  pinMode(HEATBULB, OUTPUT); //HeatBulb
  digitalWrite(HEATBULB, OFF);

  pinMode(sensorPin, INPUT); //Fan
  pinMode(motorPin, OUTPUT);
```

AllTogether

```
    {
      digitalWrite(HEATBULB, OFF);
    }
  }

  else
  {
    if (t < MINTemp)
    {
      digitalWrite(HEATBULB, ON);
    }
  }


int sensorValue = analogRead(sensorPin);
float voltage = sensorValue*(5000/1024.0);
if(voltage == 0)
{
  Serial.println("Fault");
}
else if(voltage< 400)
{
  Serial.println("preheating");
}
else
{
  int voltage_diference=voltage-400;
  float concentration=voltage_diference*50.0/16.0;

  Serial.print(concentration);
  Serial.println("ppm");
}

delay(9600);

//read sensor value and set upper limit cap
  sensorVal = analogRead(sensorPin);
  if(sensorVal > 4000){
     sensorVal = 4000;
  }
  //map and assign pwm values to the fan output 0 to 255 corresponds to 0 to 100%
  motorPinVal = 255;

  // the lower PWM limit
  if(sensorVal <2000){
    motorPinVal = 0;
  }

  //write the PWM value to the pwm output pin

//distance sensor
//digitalWrite(trigPin, LOW);
//delayMicroseconds(2);
//digitalWrite(trigPin, HIGH);
//delayMicroseconds(10);
//digitalWrite(trigPin, LOW);

  // Measure the response from the HC-SR04 Echo Pin

  duration = pulseIn(echoPin, HIGH);

  // Determine distance from duration
  // Use 343 metres per second as speed of sound

  distance = (duration / 2) * 0.0343;

  // Send results to Serial Monitor

  Serial.print("Distance = ");
  if (distance >= 400 || distance <= 2) {
     Serial.println("Out of range");
  }
  else {
    Serial.print(distance);
    Serial.println(" cm");
    delay(1000);
  }
  delay(100);
}
```

4. Publications and Valorisations

The process of biodesign and biofabrication follows a cyclical workflow, allowing for the revisiting of different stages based on findings and new insights. Generally, the process begins with concept development, which may be driven by addressing a real-life application, exploring a research question, or testing a theoretical idea. This phase often includes preliminary material testing, focusing on understanding the living material, cultivating it, and experimenting with its potential. Once the material demonstrates promising signs of meeting the concept's requirements, the proof-of-concept phase advances to more rigorous testing of the idea.

After the preliminary tests, the framework can evolve in various directions, including the biofabrication of materials, performance measurements, post-production methods, and public engagement, depending on the project's or researcher's objectives. The specific path forward is shaped by the researcher's goals, which may range from developing commercial products to exploring agential methodologies. These approaches exist on a spectrum, balancing practical applications with the advancement of innovative biofabrication techniques. As the field continues to evolve, additional elements and considerations will likely be integrated into this framework.

In the second year of my PhD, our team secured Research England's E3 (Expanding Excellence in England) Fund to establish the Hub for Biotechnology in the Built Environment (HBBE). I was employed by the HBBE for over a year, where I engaged in various stages of the biodesign and biofabrication process. This included developing fabrication techniques and creating prototypes for exhibitions. These projects often required iterative cycles of material testing and monitoring at every stage of their development. The prototypes I worked on include:

BioKnit: This project utilized mycopaste within a knitted scaffold to create a structural form. Two versions were developed using the same methodology. The initial prototype was exhibited at the OME, while the second iteration was showcased at the London Design Museum. My contributions included concept development and conducting preliminary material tests for the first prototype, as well as building the second iteration.

The Living Room: This project evolved from the mycopaste developed for the BioKnit projects, adapting the original recipe to incorporate waste materials. Mycopaste was plastered to a knitted wool formwork to create the structure. My role included initial material testing, optimizing the paste recipe to use entirely waste-based ingredients, constructing the prototype, overseeing post-production monitoring, and testing the mechanical properties of the mycelium composite materials.

The Snout: This project focused on bacterial cellulose (BC) materials, specifically exploring methods for the intermediate stage between BC production and its application.

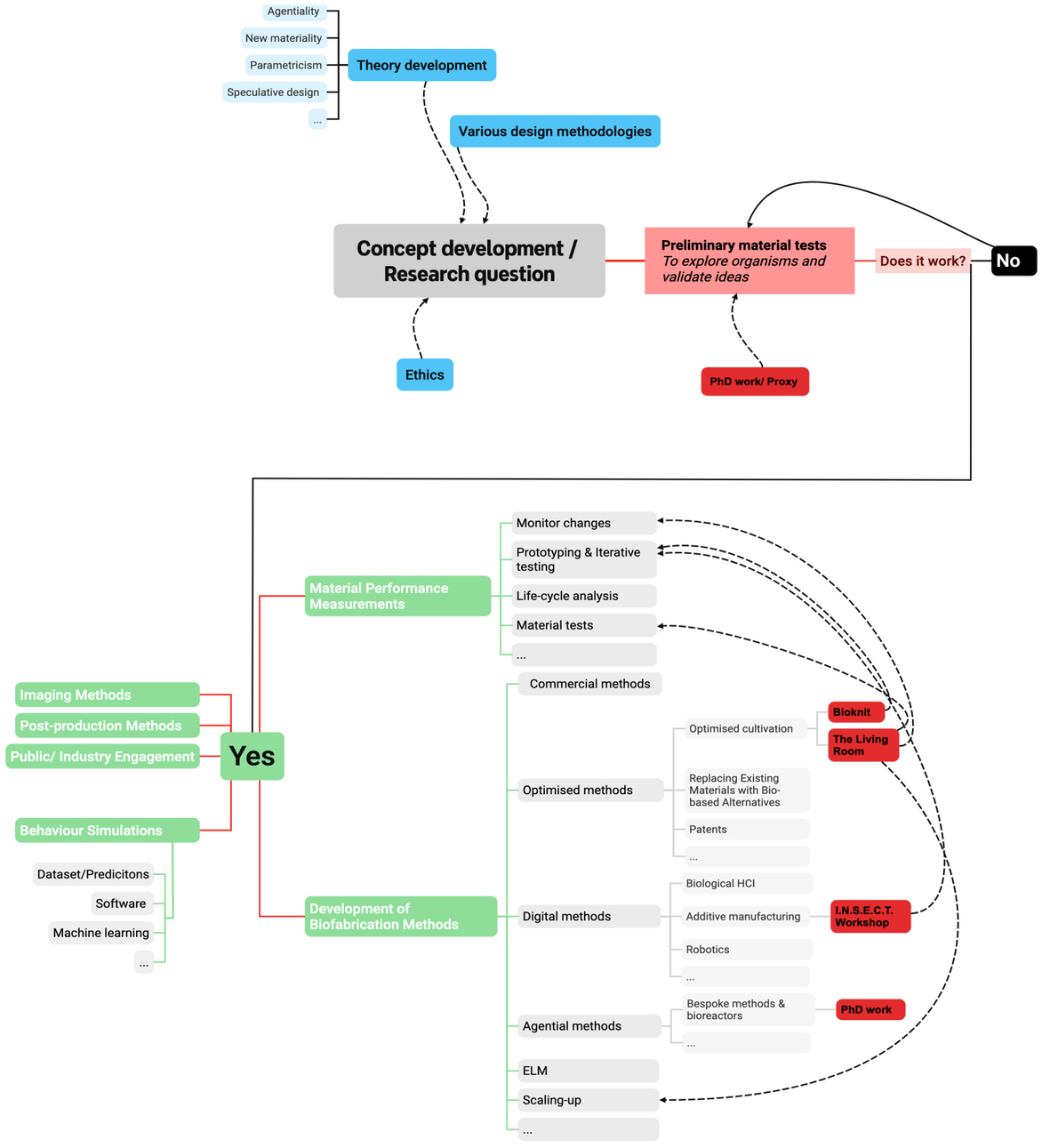

Figure 1

The framework representing various components of the biodesign framework.

OUTPUTS / PUBLICATIONS

Book Chapters

PhD Research

D. Ozkan, Designing Mushrooms, in Bioprotopia, Prototyping biotechnologies and biocultures for the built environment, Birkhäuser, 2023 (p.170-175).

D. Ozkan, Case 6 - Demonstrating a Material Making Process Through the Cultivation of Fungal Growth, in Pedgley, O., Rognoli, V., and Karana, E. (eds) Materials Experience 2: Expanding Territories of Materials and Design. Oxford: Butterworth-Heinemann, 2021.

Bioknit/The Living Room

J. Scott, B. Bridgens, D. Ozkan, R. Kaiser, The Living Room: New Expressions of Biohybrid Textile Architecture, Fabricate 2024, DOI: 10.2307/jj.11374766.8

J. Scott, E. Elsacker, B. Bridgens, R. Kaiser, D. Ozkan, A. Hoenerloh, et al., Hi-tech/ Low-tech/ Bio-tech Crafting the Bioknit Portotype, Birkhäuser, 2023 (p.61-75).

J. Scott, R. Kaiser, D. Ozkan, A. Hoenerloh, et al., Knitted Cultivation: Textiling a Multi-Kingdom Bio Architecture, in Structures & Architecture Volume 2, CRC Press, 2022 (p.3-10). ISBN: 978-0-367-90281-0.

Journal Articles

PhD Research

D. Ozkan, M. Dade-Robertson, R. Morrow, M. Zhang, Are mushrooms parametric? in Biomimetics Special Issue, Fungal architectures, 2022.

A. Hoenerloh, D. Ozkan, J. Scott, Multi-organism composites: Combined growth potential of mycelium and bacterial cellulose in Biomimetics Special Issue, Fungal Architectures, 2022.

Conference Proceedings

PhD Research

D. Ozkan, M. Dade-Robertson, R. Morrow, M. Zhang, Fungal Forms: Utilising Developmental Plasticity as a Biofabrication Strategy, in ICSA, 2025.

D. Ozkan, M. Dade-Robertson, R. Morrow, M. Zhang, Designing A Living Material Through Bio-Digital Fabrication, in eCAADe Towards a New Configurable Architecture, 2021.

D. Ozkan, M. Dade-Robertson, B. Christgen, Demonstrating a Material Making Process Through the Cultivation of Mycelium Growth, in (BIODIG) 4th International Conference for Biodigital Architecture & Genetics, 2020.

Bioknit/The Living Room

J. Scott, B. Bridgens, R. Keiser, D. Ozkan, et al., THE LIVING ROOM: Interaction of physical making and digital modelling for large scale biofabrication, in ACADIA Habits of the Anthropocene, 2023.

J. Scott, B. Bridgens, D. Ozkan, et al., BIOKNIT-TWO: Transitioning to Regional Waste Streams for Biohybrid Textile Production, in the 2nd international conference on Construction, Energy, Environment &Sustainability, 2023.

J. Scott, D. Ozkan, A. Hoenerloh et al., BIOKNIT Buildings: Strategies for Living Textile Architectures, in the 1st international conference on Construction, Energy, Environment &Sustainability, 2021.

Awards / Funding

PhD Research

2019 The Sonoma County Mycological Association-Herbert M. Saylor Memorial Scholarship ($3,000)

2019 Newcastle University Overseas Research Scholarship (approximately £11,490 per annum)

Exhibitions

PhD Research

11/2023 Fungi Entangalment, Air Space in St Petersburg, Russia

05/2021 Living Construction: Can We Grow a City?, In Venice Biennale 2021, Virtual Event, Italy
In collaboration with Martyn Dade-Robertson, Meng Zhang, Thora Arnardottir, Sunbin Lee, Emily Birch, Monika Lipinska, Ahmet Topcu

09/2019 Yggdrasil, London Design Festival at Open Cell, UK
In collaboration with Assia Stefanova, Thora Arnardottir, Sunbin Lee

Bioknit/The Living Room

04/2024 Edinburgh Science Festival
Growing Home Exhibition at the National Museum of Scotland, UK
In collaboration with Jane Scott, Romy Kaiser, Ben Bridges, Armand Agraviador, Oliver Perry

11/2023 The Bioknit-Arch
Future Observatory Exhibition at the Design Museum in London, UK
In collaboration with Jane Scott, Romy Kaiser, Ben Bridges, Armand Agraviador, Oliver Perry

04/2023 The Living Room
More with Less Exhibition at the Farrell Centre in Newcastle upon Tyne, UK
In collaboration with Jane Scott, Romy Kaiser, Ben Bridges

01/2022 Bioknit Prototype
In the OME in Newcastle upon Tyne, UK
In collaboration with Jane Scott, Aileen Hoenerloh, Romy Kaiser, Armand Agraviador, Ben Bridges, Ahmet Topcu, Elise Elsacker

Lectures / Public Talks

12/2023 “Fungi Entanglement” at the Art & Science Centre of ITMO University

09/2022 “Digital Fabrication of Biomaterials” in Fraunhofer AICOS at the Thursdays with Science session

04/2022 “Living Artifacts for Daily Life” for interior design students at MEF University

04/2021 “Computational Based Design” for the first-year Basic Design students at Bilgi University

09/2020 “Prototyping the Future of Biomaterials” in Bio Art & Design track at the Bio Summit Conference

06/2020 “Growing Artefacts” at the ARCHINTEX Conference hosted by the Swedish School of Textiles

Workshops

10/2020 In Silico, In Vitro, In Vivo: Programming in Processing and in Life Workshop, USA, ACADIA Conference Workshop-09
In collaboration with Martyn Dade-Robertson, Carolina Ramirez Figueroa, Jane Scott

01/2020 Design Technologies’ module, and Linked Research module, Stage 5, at Newcastle University
In collaboration with Martyn Dade-Robertson and Thora Arnardottir

The HBBE focused on four core themes: Building Metabolism, Living Construction, Microbial Environments, and Responsible Interaction. Additionally, it supported several Special Interest Groups (SIGs), which provided platforms for fostering collaboration and exchange across multiple themes, representing emerging areas of interest within the HBBE.
I established one of these SIGs, Mycology for Architecture, as an international collaboration for designers working with fungi as a biomaterial. Within this SIG, I organised seven online talks by inviting three to four experts on the topic covering areas such as:

1. 3D printing mycelium composites
2. Mycelium structures
3. Big scale applications of mycelium composites
4. Fungal leather
5. Theory & methodology of working with fungal materials
6. Fungi in collaborations with other organisms

In addition to these talks, I received a grant from Connected Everything to fund a workshop titled “Interspecies Exploration Through Bio-Digital Manufacturing Technologies.” During this nine-day workshop, with nine participants and three other co-leads (Asya Ilgun, Svenja Keunen and Laurin Kilbert), we explored clay 3D printing, living mycelium, and textiles to promote the colonization of insects and microorganisms.

BOOKS

D. Ozkan (editor), Mycology for Architecture, Pelagic Publishers, Spring, 2026.

ORGANISED EVENTS

2022 Co-organiser in the Interspecies Exploration by Bio-Digital Manufacturing Technologies (I.N.S.E.C.T.) Summer Camp Part-1 at Newcastle University
In collaboration with Asya Ilgun, Svenja Keune, Laurin Kilbert, Artificial Life Fab, HBBE, FabLab Barcelona, Mycology for Architecture

AWARDS/ FUNDING

2022 The Danish Arts Foundation, Craft and design projects in Denmark and abroad funding, for the I.N.S.E.C.T. Summer Camp (€10,000)
In collaboration with Asya Ilgun, Svenja Keune, Laurin Kilbert

2022 Connected Everything, Events host funding for the I.N.S.E.C.T. Summer Camp (£2,500)