Enacting synthetic life: Between the biological and the electronic in the Design-Build-Test-Learn bioengineering workflow

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Contents

Abstract

This thesis aims to examine how fully synthetic organisms are materially crafted in the "Design-Build-Test-Learn" bioengineering workflow, which has emerged in recent years at the intersection of synthetic biology, metabolic engineering, and computer science. To carry out this project, I analyse published scientific articles documenting actual deployment cases of Design-Build-Test-Learn pipelines, focusing on the process of creating entirely artificial biological matter as well as on the design specifications guiding this. The analysis produced two main results. Firstly, biological matter comes to life through the communication of two distinct environments (ontologies), the laboratory of 'wet' biology and the electronic computer, with a multiplicity of technoscientific objects, inscriptions, and translations mediating the process. Secondly, the design choices involved inscribe a hybrid digital logic to the outcome of the workflow, the artificial single-cell organism. This thesis could be useful for science and technology studies (STS) scholars since it contributes: a) A micro-level material analysis in bioengineering discussions, arguing that the current literature regarding the role of metaphors and ethical responsibility fails to account how discursive practices and ethical considerations interact with the design script of bioengineered entities; b) To the ontological approaches in the field, by demonstrating the versatility of such a framework, especially in highlighting the political stakes involved in bioengineering practices. This thesis could be also useful for bioengineers because it invites them to reconsider the current narrative of problem-solving in synthetic biology, reflecting on the input–output framing of societal problems and the corresponding engineering solutions developed in a research-for-industry context.

1 Introduction

In this introductory section I firstly set the thematic background of the thesis, presenting the field of synthetic biology and the corresponding type of laboratory that has emerged alongside it, the biofoundry. Then, I demarcate the object of my research, namely the "Design-Build-Test-Learn" bioengineering workflow that is the organisational framework in biofoundries and the engine driving synthetic biology. Finally, I provide an overview of the thesis structure, briefly presenting my approach.

1.1 Setting the stage: Synthetic biology and biofoundries

In 2012, the inaugural editorial of the *ACS Synthetic Biology* journal by the American Chemical Society provides a general description of the research area with the same name that had been gradually developing for a few years already by then:

> Synthetic biology aims to improve the process of genetic engineering. It looks to a future where the design of genetic systems and the idiosyncrasies of DNA are decoupled, and one can compose living systems by mixing-and-matching genetic parts. (Voigt, 2012, p. 1)

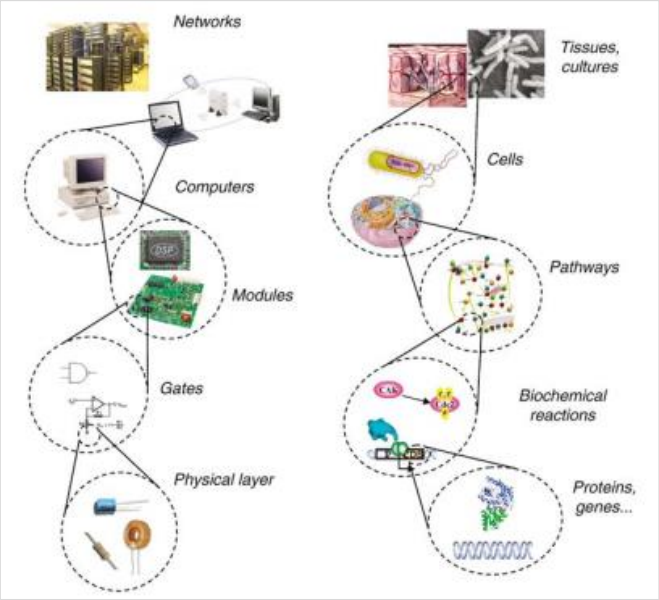
The next few lines of the text emphasise the interdisciplinarity of the nascent field, mentioning that both engineering ("biological, chemical, and electrical") and basic science disciplines ("chemistry, biology, mathematics, and biophysics") will need to join forces for synthetic biology to realise its full potential. Three years later, the inaugural editorial of the *Synthetic Biology* journal (published by Oxford University Press) embraced an evidently more promissory discourse to describe the field, calling it a "cyber-biological revolution", akin to "a cultural revolution that will have far reaching implications for the biotechnology industry", even going as far as to crown it the catalyst for an upcoming "5th industrial revolution" (Peccoud, 2016, p. 1).

Regardless of the tone it is presented in, synthetic biology is a highly interdisciplinary field at the crossroads of the life sciences and engineering; it essentially aims to turn biology into an entirely engineering discipline, so much that the manufacturing of a biological part would not be any different from manufacturing a car. It is telling of the far-reaching role engineering plays in synthetic biology that field practitioners describe older attempts at genetic engineering as throwing into a river "a bunch of concrete and

steel […] and if you can walk across, call it a bridge" (anonymous synthetic biologist as quoted in Calvert, 2013, p. 407).

Synthetic biology is inspired by a very specific kind of engineering—namely, computing engineering; hence the framing of synthetic biology as a "cultural revolution", the DNA as "the new silicon", and synthetic biologists as being "just like [the computer engineering] visionaries in the 70s" (Peccoud, 2016, p. 1). In fact, it is not unusual at all to find comparisons between the biological and the electronic in the synthetic biology literature, like the one presented below:

Figure 1: "A possible hierarchy for synthetic biology is inspired by computer engineering", as appears in Andrianantoandro, Basu, Karig, and Weiss, 2006, p. 2.



The diverging tone of the two inaugural editorials I quoted above is telling of the constitutive narratives of synthetic biology. On the one hand, there is the scientific practice, which is unintelligible outside of the bioeconomy and especially without its foundational connection with the energy, pharmaceutical, and chemical industries. On the other hand, there is the promise that synthetic biology will be instrumental in solving social problems. The conjuring of both tones can be observed in the "Synthetic Biology Roadmap for the UK", one of the earliest policy documents of its kind:

> Synthetic biology is the design and engineering of biologically based parts, novel devices and systems as well as the re-design of existing,

> natural biological systems. The step change in the synthetic biology approach is to engineer biological systems to perform new functions in a modular, reliable and predictable way, allowing modules to be reused in different contexts. It has the potential to deliver important new applications and improve existing industrial processes across many sectors including healthcare, energy, pharmaceuticals, materials, and remediation – resulting in economic growth and job creation. (Synthetic Biology Roadmap Coordination Group, 2012, p. 12)

The two voices of synthetic biology, however, are part of different sites and pathways of knowledge production. The promissory one is mostly reserved for the "policy room" (Marris and Calvert, 2020), while the scientific one for the new type of laboratory that co-emerged with synthetic biology, the biofoundry[1]. Although I will touch upon the promissory voice during the literature review and comment on the interrelation of the laboratory and the policy room in the conclusion, the focus of my research will be put on the scientific voice and its corresponding pathway.

Synthetic biology cannot and should not be thought separately from the site where it is practiced, the biofoundry, which has been described as a factory for genetic products (Freemont, Curach, Friedman, and Lee, 2019). This type of laboratory is tailored to the high-throughput and advanced technological requirements of synthetic biology:

> A biofoundry is an integrated molecular biology facility that includes robotic liquid-handling equipment, high-throughput analytical equipment, and the software, personnel and data management systems required to run the equipment and broader biofoundry capabilities. (Holowko, Frow, Reid, Rourke, and Vickers, 2020, p. 1).

Biofoundries are essentially the upper echelon of biological laboratories, commanding a large array of resources in terms of funding received, technological abilities, and

[1] A note in passing: In the primary sources I studied (see the "Methodological approach" section), the promissory tone was reduced to scarce appearances in the introduction of scientific publications, which were almost identical in content across all texts.

trained personnel. The heavy use and integration of multi-level automation, from mechanical (liquid-handling robotic platforms) to computational (bespoke software and machine learning tools), situates biofoundries "at the forefront of a paradigm shift in biological engineering toward a more automated, design-focused venture" (Holowko, Frow, Reid, Rourke, and Vickers, 2020, p. 2). For example, the Agile BioFoundry, established in 2016 (which is probably the largest of its kind), is funded by the U.S. Department of Energy's Bioenergy Technologies Office and functions in a distributed model, drawing from the long history and top-tier capacities of a consortium of national and already well-established large laboratories[2].

1.2 Demarcating the object of research

The engine moving biofoundries is the so-called "Design-Build-Test-Learn" (DBTL) workflow, which acts as the organisational core of synthetic biology experiments conducted in such laboratories (Hammang and Frow, 2019). This workflow—sometimes called framework or pipeline or cycle (to denote only one iteration of the DBTL)—functions as the orchestrating pattern that orders all the various infrastructural elements and resources involved in a biofoundry. Appropriating the way engineers traditionally frame problem-solving (Hammang and Frow, 2019), the DBTL workflow:

> [A]ims to fulfill particular design criteria for a synthetic biology application, which might for example be the production of a specific product at an optimal titer or the detection of a specific clinical biomarker using an engineered gut microbiome. (Hillson et al., 2019, p. 2).

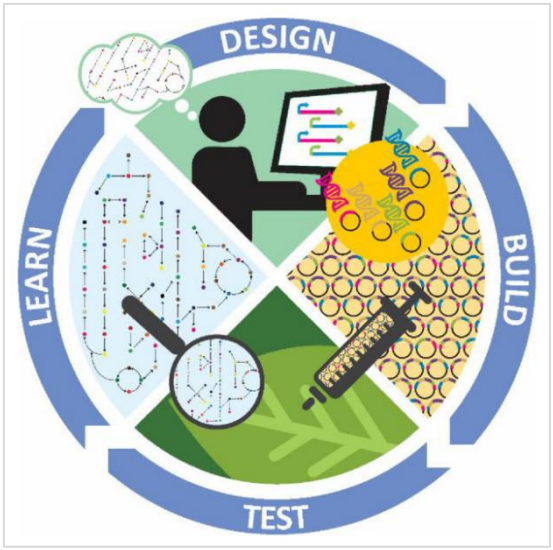
The DBTL workflow, in simple terms, is the iterative step-by-step bioengineering methodology used by synthetic biologists to rationally optimise and/or design anew a metabolic pathway/biosystem, place it in a host (this is usually carried out by manipulating the DNA of Escherichia coli bacteria—the gut microbiome above) and thus create an entirely synthetic single-cell organism that produces in increased volume (the optimal titer and the detection mentioned above) a certain substance of interest

[2] These are: Argonne National Laboratory, Lawrence Berkeley National Laboratory, Los Alamos National Laboratory, National Renewable Energy Laboratory, Oak Ridge National Laboratory, Pacific Northwest National Laboratory, Sandia National Laboratories.

which is not native to the host. For example, a DBTL experiment conducted in the SYNBIOCHEM biofoundry (in Manchester, UK) designed synthetic DNA and inserted it in multiple E. coli hosts with the goal of increasing the production of various monomers (e.g., cinnamic acid, which is used in the manufacturing process of various cosmetic, food, and pharmaceutical products) (Robinson et al., 2020).

The DBTL workflow has four parts, as its name indicates. During the Design phase, synthetic biologists choose the biological pathway to be engineered or modified, the target substance to be monitored, and the appropriate host to be used. In the Build phase, these building blocks are synthesised and transferred to the selected host, while during the Test phase the organism that has been built undergoes various analytical screenings to determine whether the design goal has been achieved. Finally, in the Learn phase, synthetic biologists assemble the data from the screenings, using it to improve their original hypothesis, and thus tinker with their initial design parameters in the next cycle of DBTL to be performed (Pouvreau, Vanhercke, and Singh, 2018). Depending on the scope of the project, a DBTL workflow may span from a few iterations/cycles up to hundreds, usually when performed in a market-oriented context. The DBTL is usually depicted in the following manner:

Figure 2: "Schematic representation of the DBTL cycle", as appears in Pouvreau, Vanhercke, and Singh, 2018, p. 4.



The figure above is indicative of the fact that the DBTL workflow is a "morality tale of efficiency" (anonymous synthetic biologist as quoted in Hammang and Frow, 2019).

The idea is that such a standardised step-by-step approach can be automated and optimised so that the production of substances of interest (e.g., for new medicine) is not as time-consuming as it currently is. It is quite common in the synthetic biology literature to start an article by contrasting the person-years (method of calculating effort) required in the past to produce a substance of interest with the person-years required to produce a substance under the DBTL workflow, emphasising that the DBTL would be even more efficient in the future. In a relevant example, the 575 person-years required by the multinational chemical company Dupont to generate propanediol, which is the base for their Sorona fabric, is contrasted with the 150 person-years required by the biotechnology company Amyris to produce artemisinin by employing multiple DBTL cycles (artemisin is the main ingredient used for manufacturing anti-malaria medication) (Radivojević, Costello, Workman, and Martin, 2020). The current sentiment in the literature is that even the effort required by Amyris is already too much, with the goal being to reduce it significantly in the following years (Gill, Halweg-Edwards, Clauset, and Way, 2016).

1.3 Brief overview of the thesis

There are many lines of inquiry that could be explored regarding the DBTL workflow. To name only a few—expanding the issues scoped by Hammang and Frow (2019)—from a science and technology studies (STS) perspective, someone could inquire about the rationality of bioengineering design, the relation of the cycle and its schematic representation with industrial organisation studies, the relation of automation to manual labour and tacit knowledge, the design values involved in the process, the commodification of DNA, and the negotiation of failures by synthetic biologists to achieve the expected results.

However, I will focus on the 'journey' of synthetic biological matter in the DBTL workflow as orchestrated at an ontic/material level through processes of inscription, translation, and scripting that take place between the four workflow phases (Design, Build, Test, Learn). To follow the journey of biomatter throughout the multiple phases of the DBTL, I have used as a prototype the way Annemarie Mol (2002) follows the various enactments of atherosclerosis in a Dutch hospital, tinkering with her approach to fit the particularities of my research object. To that end, following John Law (2004), I have paired the concept of inscription (Latour, 1987) with that of enactment; in

addition, I have selectively drawn from the concepts of translation (Latour, 1994) and script (Akrich, 1992) to further refine what a chain of enactments would mean in the context of the DBTL. I explain how all these fit together in the "Theoretical framework" section of the thesis, while in the "Research question" section I outline what I wanted to study, phrased in the vocabulary of my theoretical framework.

The methodological idea of a technoscientific object travelling across contexts is borrowed by Lionelli (2014) and fits nicely with Mol's approach—although Lionelli employs it for data while I use it for biomatter. By zooming in on specific transitive moments of biomatter in the DBTL as described in published texts that depict DBTL experiments (i.e., when biomatter passes from one phase to the next), I demarcate snapshots of the journey that I then analyse to describe the way biomatter is crafted, with its divergences and unifying logic. Overall, my aim is "to keep track as persistently as possible of what it is that alters when matters, terms, and aims travel from one place to another" (Mol, 2002, p. viii). I outline in detail my methodological approach and choices in the "Methodological approach" section.

In the "Analysis" section I discuss and analyse in length three snapshots from the DBTL workflow: The passing of biomatter from Build to Test (in the "Decontextualization" section of the thesis), from Test to Learn (in the "Recontextualisation" section), and from Learn to Design (in the "Reuse" section), concluding the tracking of biomatter through a full DBTL cycle. In each snapshot, I identified that biomatter oscillates between two distinct ontologies: The ontology of wet laboratory biology and the digital one of the electronic computer. It is this circular oscillation that sets in motion processes of inscription, translation, and scripting in the workflow, through which distinct ontic states of biomatter are enacted: From biological it becomes electronic (data); from data it changes into a visual form (picture); from being visual it assumes an architectural structure (order of DNA code), before finally turning again into its biological form once again. By doing this, I argue that there is an ontological script that acts as a unifying equaliser between the various enactments of biomatter, materially inscribing the binary logic of an electronic circuit into biosystem functions of the single-cell organism. Thus, I conclude that the design choices involved in the DBTL are the result of the ontological choreography of enactments taking place when the wet biology ontology and the

ontology of the electronic computer are summoned to interact and become mutually intelligible within the DBTL workflow.

Before all that, in the "Literature review" section of the thesis, I review the STS literature on bioengineering and synthetic biology, as the Design-Build-Test-Learn workflow falls within their scope. To my knowledge, this workflow has not so far been discussed in the secondary literature, despite a handful of published works discussing some of its elements. By surveying thirty-one (31) articles and books of the relevant STS literature on synthetic biology, I identify a lack in approaches that pay attention to its infrastructure and practices, in particular as concerns synthetic biology workflows. The main takeaway from the literature review is that social and ethical considerations dominate the discussion, coming out of a technology governance perspective or a discursive one focusing on machine metaphor use, while there is a lack of cases that inquire into the material specificities of synthetic biology.

2 Literature review

2.1 Methodology for literature collection

In this section, I review the STS literature on bioengineering and synthetic biology. To collect the literature, I focused on articles from STS journals and books that mentioned synthetic biology and/or one of the following subjects: Metabolic engineering (which is a sub-type of synthetic biology prevalent in the DBTL), engineering in biology, biofoundries, bioengineering workflows, Design-Build-Test-Learn/DBTL. At first, I examined the following journals that are heavily referenced in the STS community: a) *Science, Technology, & Human Values*, b) *Social Studies of Science*, c) *Science, Technology and Society*, and d) *Technology and Culture*. After I identified multiple articles, I went through their references through a snowballing method, ending up with twenty-nine (29) articles and two (2) books, which are discussed below.

I did not extend my literature survey to neighbouring terms (for example, the Human Genome Project or genetic engineering in general) for three reasons. Firstly, although synthetic biology stands at the end of a long path of biology-engineering mediations, I wanted to grasp the specific debates that exist on synthetic biology and not historically between engineering and biology in general. Secondly, synthetic biology is a distinct field with distinct tools and approaches. Thus, debates from neighbouring fields did not seem entirely transferable to the particularities of synthetic biology (for example, see my discussion of Keller [2002] below). Thirdly, opening up the literature review to the long line of historical mediations between biology and engineering would not be possible in the context of this thesis, due to the limited scope of a master's thesis.

2.2 The four camps identified

Four thematic camps were identified in STS, according to their approach towards synthetic biology: a) "Constructing nature", b) "post-ELSI governance", c) "metaphors", and d) "infrastructure and practices". Each one will be explained in detail below. A Table at the end of the section provides an overview of the four camps and the literature items falling under each camp.

Summarising the literature review lessons learnt, one observation can be made, and one gap can be identified. Regarding the observation, based on the volume of contributions, the second camp (post-ELSI governance) is the largest in terms of volume; the smallest

is by far the fourth one (infrastructure and practices). If the third camp (metaphors) is added to the second, due to their common concerns, then it is obvious that the issues of the social and ethical framing of synthetic biology are the ones dominating the discussions of synthetic biology in STS.

Regarding the gap identified, it was striking that there were very scarce inquiries into the materialities of the workflows of synthetic biology. In fact, there is no mention of the Design-Build-Test-Learn workflow apart from a passing mention in Hammang and Frow (2019). This is somewhat surprising, considering that the synthetic biology literature is adamant that DBTL is the engine behind the biofoundries that drive synthetic biology forward.

As I will elaborate further in the "Methodological approach" section, the main reason for this could be the very recent public emergence of biofoundries and their workflows. For example, the Global Biofoundries Alliance, an alliance of public biofoundries around the world, was inaugurated right before the COVID-19 pandemic occurred, in December 2019 (Hillson et al., 2019). In addition, it seems that most biofoundries were established and the term gained traction after 2015. Finally, the results of the first DBTL experiments did take some time to reach the public through publications, with the first scientific publications explicitly mentioning the DBTL workflow dating around that time (e.g., Nielsen and Keasling, 2016).

2.2.1 The first camp: Constructing nature

Literature belonging to the first camp is looking into the conceptual implications of introducing engineering into biology, primarily tackling the issue either from a historical or a social perspective. The overarching question driving this camp is: What does it mean to construct nature? Here, the works of Keller (2009a, 2009b) and Calvert (2010, 2013) feature prominently.

Keller, echoing the standpoint of Rheinberger (2000), argues that synthetic biology has fundamentally shifted biology from a discipline of understanding nature into a discipline of intervening and remaking it, asking about the implications of this regarding the reproduction of organisms. However, she mentions (in line with Campos [2009]) that both standpoints were there from the early days of biology—for example, when Jacques Loeb was talking about synthetic biology and wanted to produce artificial

life in the early 20th century. Along these lines, Keller (2002) has also grappled with the implications of constructing synthetic organisms from a historical standpoint, situating artificial life under the long history of automata in science; however, she has focused on computer modelling methods that do not feature prominently in the field of synthetic biology.

Calvert has had a continuous and multifaceted engagement with the field of synthetic biology, from its early days in the 2000s up to now, demonstrating issues that are worth questioning within synthetic biology from an STS perspective. She has shown how the construction of nature is taking place along the lines of Rabinow's concept of "biosociality" (Rabinow, 2005; Calvert, 2010) and how synthetic biologists could be very well understood as a paradigmatic type of heterogeneous engineers (Calvert, 2013). Continuing this thread of research, Frow and Calvert (2012) have investigated how teams in the established synthetic biology competition "iGEM"[3] negotiate with design choices (when constructing a synthetic organism) that are more complex than the engineering rhetoric would imply. In a follow-up article, Schyfter, Frow, and Calvert (2013) have explored the overall connection of synthetic biology with engineering, and Frow (2013) has explored how value is created in synthetic biology.

2.2.2 The second camp: Post-ELSI governance

The approaches belonging to the second camp investigate issues of governance, drawing mainly from "responsible research and innovation" (RRI) paradigms in a post-ELSI ("ethical, legal, and social implications") context. Such approaches focus mainly on the level of transparency and the level of public involvement in the decision-making process in synthetic biology. The overarching question connecting this camp is: How can the innovations in synthetic biology be shaped according to ethical and social concerns through public and transparent deliberations?

All approaches in this camp aim to overcome the problems associated with the downstream-style engagement framework of ELSI (Balmer et al., 2016), as the

[3] iGEM is the "International Genetically Engineered Machine" competition taking place each year since 2004 in Boston, U.S.A and involves mainly undergradutates teams (more recently it included high school students and other non-student groups) that compete for the best synthetic biology construct. It has consistently grown every year, reaching 353 teams as contestants in 2019.

governance of synthetic biology is based on inadequate governance models (Jasanoff, Hurlbut, and Saha, 2015). In this light, the RRI has emerged as a strong paradigm from which STS researchers draw inspiration. It is worth mentioning that RRI, as a governance framework stipulating that technological innovation will be shaped towards social goals, has been adopted as the European Union's public engagement tool under its research programmes, featuring prominently, for example, in the "Horizon 2020" one (de Saille 2015).

In this post-ELSI space influenced by RRI, scholars aim to identify how synthetic biology innovations can be responsive to social concerns, working alongside stakeholders and the public and overcoming disciplinary and institutional limitations encountered (Torgersen and Schmidt, 2013; McLeod, Brigitte Nerlich, and Mohr, 2017; Delborne, Kokotovich, and Lunshof, 2020). The manner that the public is engaged in synthetic biology (in its strategic direction and policymaking aspects) is of crucial importance to scholars in this camp, especially the terms that the public debate (Meyer 2017) and the relevant groups (Frow 2020) are constructed. In a relevant case study, Balmer and Bulpin (2013) show that a component on 'human practices' (i.e., regarding social implications) that exists in some synthetic biology research groups are narrowly defined based on the ELSI framework.

Within this camp, there are a few approaches that deal with the way STS researchers in particular negotiate their role in synthetic biology projects. Two prominent examples were surveyed, one outlining the engagement with the first public-funded synthetic biology project in the USA (Rabinow and Bennet, 2012) and the second one outlining the engagement with the working group that defined the 2012 UK policy roadmap on synthetic biology (Marris and Calvert, 2020). Both approaches highlight the limited space left to social scientists to define the terms of engagement, which have been already predefined in a narrow ELSI framework (at best). In fact, in the case of Rabinow and Bennet, the engagement ended up in an eventual split-up with the project and their substitution.

2.2.3 The third camp: Metaphors

The third camp in the relevant STS literature includes approaches that focus on the metaphorical use of language in synthetic biology and the ethical and social

implications of this. This camp is essentially the other side of the second camp because both inquire about social and ethical issues but from a different perspective (second camp: Deliberation processes; third camp: Use of language). The overarching question driving the third camp is: What are the moral and social implications of the heavy use of metaphors in synthetic biology?

This camp is concerned with issues emerging from the metaphorical use of language in synthetic biology. The main element of concern that scholars adopting this approach outline is the power of metaphors to frame and entrench a life-as-machine paradigm in biology that renders unproblematic the notion of engineering living organisms (Kearnes, Kuch, and Johnston, 2018; Grote, 2019). For this reason, the analogy of human-made machines with molecular processes may be inadequate to describe the complexity of the latter (Boudry and Pigliucci, 2013), even having implications for how the public is invited to engage with synthetic biology (O'Keefe et al., 2015). Thus, it is both a) imperative to critique the application of machine metaphors (Boldt, 2018) or b) use them responsibly in line with the moral implications they already have (Deplazes-Zemp, 2012; McLeod and Nerlich, 2017).

2.2.4 The fourth camp: Infrastructure and practices

The fourth camp, which is by far the one with the fewest interlocutors involved, is concerned with questions of infrastructure and practices in synthetic biology, in particular in terms of automation. The overarching question driving this camp is: What is the impact of increased automation in the practices of synthetic biology? Here, there is only one article (Meckin, 2020) and one report generated in the context of the EU-funded project (Hammang and Frow, 2019). In that article, Meckin shows how the introduction of highly automated functions by robotic handlers in strain development produces a mismatch between the tacit knowledge of biologists and processes of automation. To that end, he argues that biologists have to negotiate with automated processes and, as a result, the work of synthetic biologists is reconfigured to comply with the automated infrastructure of biofoundries.

Hammang and Frow (2019) present the results of a collaborative workshop conducted in the context of the EU-funded project “ENLIFE – Engineering life: Ideas, practices

and promises"[4] between synthetic biologists that worked in public biofoundries or biotechnology companies and STS researchers. The workshop revolved around the mapping of synthetic biology workflows and the variety of issues that emerge from reflecting on them, such as their diversity and complexity, values driving the engineering design, failures and overall rationality, the particularities of engineering in biology, and workflow as a tool for reflection.

Finally, it is worth mentioning the research conducted by Balmer, Bulpin, and Molyneux-Hodgson (2016), which employed the theoretical framework of enactment to study a synthetic biology project in the UK water industry. Although it appears quite close to my research, their investigation of ontologies stays mostly on the level of everyday interaction between relevant groups (scientists, industry players, STS researchers), without going into much detail about the material processes of creating synthetic matter in the laboratory. For this reason, however, my research could be viewed to be complementary to theirs, providing a case study of the backstage of their investigation.

[4] The project was running between 2014 and 2019 and was funded by the European Research Centre under the Seventh Framework Programme (a European Union research and development funding programme) with Jane Calvert of the University of Edinburgh as the principal investigator. All information regarding the project are available in the CORDIS website of the European Commission: https://cordis.europa.eu/project/id/616510.

Table 1: Literature review; items have been grouped into four distinct camps in chronological order

Overview of literature surveyed (total count: 31)			
Group 1: Conceptual issues in synthetic biology (10)	**Group 2: Post-ELSI governance (11)**	**Group 3: The implications of machine metaphors (7)**	**Group 4: Infrastructure, practices, and workflows (3)**
Rheinberger, 2000	Rabinow and Bennet, 2012	Deplazes-Zemp, 2012	Balmer, Bulpin, and Molyneux-Hodgson, 2016
Keller, 2002	Balmer and Bulpin, 2013	Boudry and Pigliucci, 2013	Hammang and Frow, 2019
Campos, 2009	Torgersen and Schmidt, 2013	O'Keefe et al., 2015	Meckin, 2020
Keller, 2009a	de Saille, 2015	McLeod and Nerlich, 2017	
Keller, 2009b	Jasanoff, Hurlbut, and Saha, 2015	Boldt, 2018	
Calvert, 2010	Balmer et al., 2016	Kearnes, Kuch, and Johnston, 2018	
Frow and Calvert, 2013	McLeod, Nerlich, and Mohr, 2017	Grote, 2019	
Calvert, 2013	Meyer, 2017		
Frow, 2013	Delborne, Kokotovich, and Lunshof, 2020		
Schyfter, Frow, and Calvert, 2013	Frow, 2020		
	Marris and Calvert, 2020		

3 Theoretical framework

This chapter presents the concepts I employ to analyse the journey of synthetic biomatter in the Design-Build-Test-Learn (DBTL) workflow and to situate my research in the broader theoretical landscape of science and technology studies. To this end, I engage with and draw from the theoretical pool of what has been debated as the *ontological turn* in STS and is sometimes called post–actor-network theory (post-ANT) (Woolgar and Lezaun, 2013).

The chapter is structured in four parts. In the beginning, I provide an outline of the main argument of the ontological turn, its implications, and introduce the concept central to it, enactment (Mol, 2002; Law, 2004). Afterwards, in the second part, I introduce three concepts to supplement enactment: a) Inscription (Latour, 1987), b) translation (Latour, 1994), and c) script (Akrich, 1992). In that part, I present the three supplementary concepts and rework them towards an ontological understanding, to enrich the overarching concept, enactment. In the third part of the chapter, I explain the reasons for choosing to employ such a constellation of concepts instead of another STS theoretical framework. In particular, I aim to explain their significance not only in the context of my research but also in the ways they bring to the fore the political stakes of how biomatter is negotiated and enacted in the bioengineering workflow of DBTL. Finally, in the fourth part of the chapter, I aim to elucidate disciplinary (mis)associations in the ontological turn debate, focusing on approaches from two of the main interlocutor-disciplines, anthropology and philosophy.

3.1 The ontological turn and the concept of enactment

In broad terms, the ontological turn in STS refers to the notion that there is a multiplicity of entities that come into being in technoscientific settings through particular arrangements of people, methods, instruments, materials, and so on (Law, 2004). There are three main implications of this. Firstly, the entities produced may compete or converge depending on the way arrangements bring them to existence, as they are being continuously (re)negotiated—affirmed or negated—through practice. That is, certain entities are perceived as stable and grouped under the same name because they have been performed in the same way and through the same arrangements countless times (for example, what is considered a scientific fact). Conversely, other entities may be

perceived as unstable and multiple names may exist for them, as they may be brought to existence through contradictory performances of arrangements.

Secondly, what could be perceived as a fixed entity or object—for example, the disease of atherosclerosis in Mol (2002)—comprises multiple and often competing objects that have been crafted through different arrangements yet have been reified under a single name. Thirdly, if reality is examined in such an ontological framework, it becomes crowded with gaps, rifts, convergences, and synergies among different objects, not all of which are visible without attention to the performances of arrangements. Crucially, there is no singular reality for which different perspectives or representations exist, but the world is revealed as potentially multiple in its very composition (hence the use of the word in plural, ontolog*ies*, often used in the relevant literature).

Enactment lies at the core of the ontological turn. Annemarie Mol, who first employed this concept in her book *The Body Multiple* (2002), follows the multiple arrangements that craft a single entity, atherosclerosis, a potentially fatal disease of the arteries, in a Dutch hospital. She attends to the various sites of production of atherosclerosis, focusing on five (yet with many more being alluded to in the book): The surgeon's consulting room, the pathology laboratory, the radiology department of the hospital, the room where ultrasound tests are carried out, and the operating room where the surgery takes place.

In each site, atherosclerosis is "framed as parts of events that occur and plays that are staged" (Mol, 2002, p. 44), since each site has its own arrangement of actors involved (human and non-human) and which sometimes produce diverging or converging objects. An angiography—a type of medical imaging technique used to show the blood flow in arteries—is produced in the radiology department and sometimes paints a quite diverging picture of arteries compared to the one crafted in the ultrasound room, where duplex techniques are used to produce an image of arteries as well (Mol, 2002, p. 83). Different object-producing sites with different arrangements of actors bring to existence atherosclerosis as two different objects. Enactment is the concept that explains this performative, continuous negotiation that multiple arrangements are engaged in and for which a singular name exists (atherosclerosis). Different arrangements bring to existence, *enact*, different technoscientific objects.

3.2 Supplementing the concept of enactment

Inscription

The concept of inscription, as elaborated by Latour, bodes well with Mol's investigation of technoscientific arrangements. In *Science in Action* (1987), Latour's skeptical reader wanted to move from the maze-like scientific text to the laboratory, where, however, they were met with a similar challenge: "We came to the laboratory in order to settle our doubts about the paper, but we have been led into a labyrinth" (Latour, 1987, p. 67). Behind the scientific text (the "let me show you" moment of the Professor in Latour's narration) were not "simple flashes of intuition" but a "slow, protracted and complicated staging of tiny images" that involved preparation of materials, tuning of machines, and calibration of more machines (Latour, 1989, pp. 67–68). The term Latour uses to denote this staging is *inscription*.

As in Mol's narration, the more attention is paid to practices, the more one is perpetually encountered with further mediation rather than the-thing-itself. The substance Latour was hoping to "see with his own eyes" (1989, p. 67) and put his doubts to rest is revealed to be a multiplicity of actors arranged in a certain manner mediating each other. Inscription, therefore, or better the process that inscription stands for, complements the concept of enactment by enriching the account of *how* a certain object emerges and is performed in laboratory settings. Inscription, then, would be the analytical tool that would allow us to see the internal workings of the enactment, the particular transformations taking place (especially when instruments and machines are involved), while an enactment would be the outcome of a certain inscription process (something is being enacted).

Translation

Mol shows how multiplicity arises through difference in the sites (and thus arrangements) of technoscientific production. Similarly, synthetic biomatter in the DBTL cycle passes through multiple sites and environments: The computer (and multiple software programmes in that respect), the growth culture, the liquid chromatographer, the mass spectrometer, and so on. The focus of Mol was on the multiplicity of objects that may be in flux behind the singular name of atherosclerosis. Yet, as the workflow of a certain DBTL cycle is ordered sequentially (the Design phase

follows the Learn phase and precedes the Build one, the Build follows the Design and precedes the Test one, and so on), the subject matter of my research forces a question regarding the communication between different arrangements and different sites of production: How do they and their objects communicate and travel from one environment to the other, from one phase to the next? The objects may be multiple and crafted through different arrangements, but Mol's account needs to be supplemented if we were to ask how one object may be substituted—or, rather, translated—from one production site to the other.

Investigating the DBTL workflow is conducive to pushing this theoretical matter further since the workflow itself is laid out in four consecutive steps. When objects from one site of production (for example, from Build) are transferred to the next (in this case, to Test), the arrangements and the objects are summoned to engage in an act of translation in order to become intelligible to each other. The name may be the same (atherosclerosis, or in my research, biomatter), but—as different arrangements are forced to communicate with each other—there is a trade-off in meaning, which is similar to how a text is translated from one language to another. There is simultaneously a surplus and a lack in the process of translation; the objects may be multiple and the sites of production different, but the DBTL workflow forces our hand to think of them as a chain of translations from one object and arrangement to another.

For this reason, the concept of translation, as elaborated by Latour, would be beneficial here. Translation is a widely used concept in actor-network theory and its meaning has changed in different phases of Latour's writings (Shiga, 2007). Here, I deploy one of the simplest definitions he gave: "I use translation to mean displacement, drift, invention, mediation, the creation of a link that did not exist before and that to some degree modifies two elements or agents" (Latour, 1994, p. 32). Taking this as the starting point, the translation of biomatter that would take place from one site of production to the other within a certain DBTL cycle exemplifies both a loss and a gain in meaning ("displacement, drift, invention, mediation" [Latour, 1994, p. 32]). Two arrangements and two objects are compelled to create a link and are forced to become intelligible to one another. This process of translation has two implications. Firstly, translation entails transformation and transmutation—i.e., one object from a certain DBTL phase morphs into a different one in the next phase. Secondly, that enactment,

as I have expanded its scope here, also involves the various translations that take place between objects summoned to communicate.

To recapitulate, enactments host a process of inscription that enables us to investigate the inner workings of how arrangements bring technoscientific entities to life. These entities, by the force of being categorised under the same name, are summoned to be translated from one environment to the other (for example, from the computer to the culture growth and from the Design phase to the Build one). Thus, different sites of technoscientific knowledge production have different processes of inscription and thus craft different objects, which are forced to engage in a mutual translation if there is a designated need for them to communicate.

Script

In the DBTL workflow biomatter is synthesised, which invites a question regarding the design of the organism to be produced in the laboratory: What is biomatter enabled and disabled to do, as performed and translated throughout a certain DBTL cycle? Since the cycle in question is an engineering approach applied to biological systems, it is worth inquiring which kind of logic biomatter is instilled with—as it has been inquired for a machine (Grint and Woolgar, 1997). If different technoscientific objects are enacted through different arrangements of actors and forced translations, then it follows that their outcomes, the single-cell organisms coming out of DBTL cycles, are designed to act in a certain way. Engineered entities, performed through certain arrangements, are materialised with specific functions and logic.

To develop this point further, I have used the concept of the script, as elaborated by Madeleine Akrich (1992). Akrich uses the term to denote that artefacts are inscribed with an instruction manual, so to say. Any artefact contains a script from its author that is addressed to the user, describing the engineered product's intended use:

> "Designers thus define actors with specific tastes, competences, motives, aspirations, political prejudices, and the rest, and they assume that morality, technology, science and economy will evolve in particular ways. A large part of the work of innovators is that of 'inscribing' this vision of (or prediction about) the world in the

> technical content of the new object. I will call the end product of this work a 'script' or a 'scenario'." (Akrich, 1992, p. 208)

Here, I narrow down the scope of the script to the level of a single-cell organism; that is the artefact in question. At the same time, I take the script to be written (to stay within the metaphor) throughout the journey of biomatter in the DBTL workflow, not limiting it to a single phase of the DBTL cycle. Therefore, the script of an engineered single-cell organism is crafted through both the inscription and, crucially, the translation process that biomatter passes through.

The concept of the script is employed here to address the promise of plurality that an ontological framework may misleadingly allude to; multiplicity, as Mol has been maintaining (1999), does not entail plurality. That is, even though arrangements are multiple, this does not entail that they produce objects that are radically different in scripts. On the contrary, the particular logic of inscription and translation converges quite well together in forming the DBTL framework. The concept of the script enables us to understand what kind of (contingent) structures multiplicity is arranged in and what its unifying logic is. As many researchers from STS have repeatedly shown, it would take a lot of effort (maybe the construction of another laboratory altogether or the establishment of different social contexts and needs) to disrupt ("de-scribe") the use scenarios of technoscientific objects (Latour, 1987; MacDonald, 2020), especially on the level of inscription.

To conclude the presentation of the quartet of concepts I employ in the context of this research, enactment acts as the overarching one and includes all others. Inscription denotes the process through which various actors are ordered together to bring to life instances of biomatter in the various phases of the DBTL workflow; translation denotes the passing from one phase (and site and inscription process) of the DBTL cycle to the next, where objects are transmuted into other objects; and script denotes the material logic instilled in the movement of biomatter through various inscription and translation processes throughout the four phases of the DBTL cycle. Thus, the inquiry into the processes of inscription, translation, and scripting is what leads us to identify the various enactments and ontological states of biomatter in the DBTL cycle. As my aim in this thesis is to examine how biomatter is—quite literally—crafted in a DBTL

workflow, the quartet of concepts I employ here allows me to attend to the specificities of this particular journey.

3.3 Reasons for choosing this framework

Fit with research scope

The conceptual framework I propose here is essentially an expanded version of Law's (2004) account of how the earlier work of actor-network theory (Latour and Woolgar, 1979) can be reinterpreted along the lines of later interventionist developments within ANT (the turn to 'ontologies', as exemplified by Mol's *The Body Multiple*). Here, I use Law's interpretation as a starting point and supplement it with the concepts of translation and script. Crucially, the framework I deploy, although it precedes my analysis of primary sources, was dictated to a large extent by the inability to use a single concept to understand the transformations of biomatter in the DBTL workflow. Three elements particular to the DBTL workflow dictated this. Firstly, DBTL has four phases, each performed in a different site and under different technoscientific arrangements. Therefore, there was a need to discern multiplicity within a unified process; this is where enactment paired with inscription comes into play. At the same time, the DBTL workflow also mandates that its multiple orderings communicate with each other; each object in each DBTL phase is understood not only as part of a chain (successive) but also acts as a substitute for all the previous ones in each new phase. This required to pair translation with enactment and inscription, situating it within an ontological framework. Thirdly, the DBTL workflow is explicitly positioned within a bioengineering paradigm; design considerations needed to be accounted for, and the concept of the script opens up my research towards that end.

Biosociality

This constellation framework is helpful for another reason. It enables me to bring to the fore in entirely material terms what Paul Rabinow has named *biosociality* (2005). By contrasting it with sociobiology—where the biological functioned in the early 20th century as a guiding metaphor for the political construction of society—biosociality appears as a concept for the 21st century, where nature is constructed based on social categories:

> "If sociobiology is culture constructed on the basis of a metaphor of nature, then in biosociality, nature will be modeled on culture understood as practice. Nature will be known and remade through technique and will finally become artificial, just as culture becomes natural." (Rabinow, 1992/2005, p. 186)

Rabinow writes in the wake of the Human Genome Project and the invention of new techniques for mapping and recombining DNA, grasping the importance of what he calls 'new genetics' for the political engineering of contemporary societies. Jane Calvert (2010) has aptly recognised this, suggesting that biosociality is a very helpful concept for understanding synthetic biology's drive to engineer nature based on computer engineering metaphors. In light of Calvert's argument, the theoretical framework I employ here enables a material understanding of how biosociality is performed, where the modelling of nature (the engineering of single-cell organisms) is based on culture (the design script of DBTL enactments). In addition, by connecting the DBTL paradigm with biosociality through an ontological framework, it becomes easier to bring into view the political stakes involved and to examine what constitutes "ontological politics" (Mol, 1999, 2002; Law, 2004; Woolgar and Lezaun, 2013, 2015).

Ontological politics

This brings me to the third reason for choosing this framework. Attending to the mechanism of difference production in sociotechnical arrangements foregrounds an interventionist and agonistic conception of doing research (ontological politics), with far-reaching implications regarding the role of STS-inclined research. Tracing the crafting of multiplicity in technoscientific settings is a way to eventualise—to institute dispute where there was none before—sites of production on the ontic, material level (and not only on an epistemic one), where artefacts—whether statistical, biological, or other—come into being and intervene. An ontological framework addresses technoscience on the level of *onto-poiesis*, i.e., attending to the production and circulation of technoscientific beings, to the political economy of enactments in technoscientific settings. Although some of the main advocates oscillate on the following issue (e.g., Mol, 1999), attending to the ontic crafting of things is less about describing reality as it is (as is the multi-naturalist case of the ontological turn in

anthropology, for example) and more about intervening in a previously self-evident circulation of inscriptions, translations, and scripts in a deflated field of action where materials, bodies, and concepts always already participate in a similar onto-poietic activity (Woolgar and Lezaun, 2015; Lynch, 2013).

Understood in this way, ontological politics has two main implications in the context of my research. Firstly, "a politics that has to do with the way in which problems are framed, bodies are shaped, and lives are pushed and pulled into one shape or another" (Mol, 1999) is attentive to the effect that framing has on the ontic level of crafting. In the case of biomatter in the DBTL, it means that such an ontological framework enables us to research the manner bioengineering is framed in the DBTL workflow and the manner biomatter is negotiated and enacted within it. Getting a bit ahead of what I will describe later on in my analysis, attending to the ontological choreography (Cussins, 1996) of the DBTL workflow shows how the crafting of biomatter is scripted with a binary logic, which reflects the binary problem-solving logic of bioengineering as an approach, where the problem (for example, fuel shortage) and its solution (for example, the production of synthetic fuel) are framed in a simple input–output way. In the case of the DBTL workflow, then, synthetic biomatter is not merely veiled in computer engineering metaphors, as a large part of the secondary STS literature correctly discusses, but it is also enacted—through a process of reducing the biologic complexity of historical biosystems and their DNA—an ontic, material level as a binary computer that computes biochemical information in an input–output format.

Secondly, ontological politics entails that research methodologies are performative gestures that intervene into the fabric of reality rather than a second-order understanding of something (Law, 2004). In other words, the way of investigating a certain issue does not only provide an account of it but essentially contributes to its production (Law, 2004, p. 5). Thus, ontological politics enables us to understand conceptual frameworks and research methodologies as both normative and onto-poietic. If technoscience intervenes in generating materially and descriptively the ontic fabric of reality, then STS-inclined research performs an equally interventionist gesture: It interferes on the level of the associations established between materials,

objects, and their descriptions (their names in Mol's terminology) and challenges them, producing different accounts of intelligibility of what technoscience is and does.[5]

Advantages over competing STS frameworks

Certainly, other STS frameworks could be applied in the context of my research. Closest to the one I employ here—and potentially equally fruitful—is the concept of heterogeneous engineering (Law, 1987) and, in general, the sociology of translation (Callon, 1986). In fact, Calvert (2013) has showed how the concept of heterogeneous engineering could be of great explanatory value to synthetic biology settings. For this reason, I would consider heterogeneous engineering a continuation of the theoretical framework I have used here. However, I chose not to use it for three reasons. Firstly, heterogeneous engineering cannot stand on its own and would still require the concepts of inscription and scripting to be applicable for investigating biomatter in the DBTL workflow, two concepts that I already use. Secondly, the fact that the DBTL workflow comprises four distinct phases demands concepts that would emphasise the multiplicity of arrangements on the ontic level, while heterogeneous engineering could be useful if the level of investigation shifted on the relations between the DBTL outcomes (synthetic single-cell organisms) and other actors beyond the laboratory. For this reason, thirdly, to incorporate the concept of heterogeneous engineering in my theoretical framework would require research that would go beyond the DBTL workflow and thus would stretch the scope of the thesis to an unsustainably long inquiry.

Two other STS theoretical frameworks could have been applied for the purposes of my research. The first is the social construction of technology (SCOT) approach (Pinch and Bijker, 1984). However, SCOT is most often employed to show how relevant social

[5] To give an example, when in *Science in Action* Latour talks about DNA, it is not just that it provides a different frame of reference from which to understand science; the account there acts upon the fabric of meaning around DNA discovery, fighting other accounts of it (e.g., accounts that would be based on *putting the phenomena first* [Pickering, 1984]). Based on this, it is obvious that the role of STS-inclined researchers is a complex one regarding technoscience. On the one hand, they put forth an antagonistic account to technoscience and its logic, which usually is some version of positivism. On the other hand, they aim to engage with technoscience in shifting not only its meaning but also its materiality (e.g., responsible innovation, etc.). Usually, either they occupy an externalist (non-inverventionist) stance—as an anthropologist, as a researcher, as a historian—or they are structurally devoid of actual inverventionist power when they aim to become internally involved (Rabinow and Bennett, 2012; see also Calvert and Marris, 2020).

groups and socially determined technical criteria are involved in gradually closing off the plasticity of an artefact. To carry out my research within such conceptual guidelines would require shifting the research focus from a snapshot-like inquiry into the ontic enactment of synthetic biomatter (more or less what I will do in my analysis) to one that would trace the shaping of biomatter through time, opening it up to other actors beyond the laboratory and essentially taking it away from inscription processes. Therefore, reorientating my focus from the ontic level to the historical one would require abandoning attention to the DBTL workflow as a site of ontic production.

The second theoretical framework that I chose not to use were policy and governance-oriented one, such as the responsible research and innovation (RRI) (Stilgoe, Owen, and Macnaghten, 2013). Although there have been many investigations of synthetic biology under such a framework in the STS literature, as I already demonstrated in Chapter 2, I consider them to be the least helpful here particularly for the following reason. Such approaches do not include conceptual tools to move the inquiry into the level of materiality. Thus, it would have been impossible to investigate biomatter in the DBTL workflow.[6]

3.4 Disciplinary (mis)associations

Let me conclude this chapter by commenting on the disciplinary (mis)associations of enactment and the ontological turn. Responding to a special issue of *Social Studies of Science*, which was thematically titled *A Turn to Ontology in Science and Technology Studies?* and guest-edited by Steve Woolgar and Javier Lezaun (2013), quite a few responses focused on the second part of the term—namely, whether there is a "turn" in STS. For example, Vasileva (2015) asked whether a "turn" is the best way to describe the attention to ontologies, Sismondo (2015) wondered whether it would be more of a smooth transition over some time rather than a turn, Aspers (2014) argued that any inquiry into ontologies in STS falls into constructivism and thus there is no need to distinguish a turn, and van Heur, Leydersdorff, and Wyatt (2012) have questioned the turn based partly on a scientometric analysis. To the extent that such responses focus

[6] The work of Lezaun (2006) is a notable exception here. Although his work has some ontological undertones regarding the emergence of GMOs as new objects of government, my research did not involve the interrelation of laws and scientific objects, thus it did not make sense to use his approach in this case.

on disciplinary categorisation ("Is there a turn or not in STS?"), it seems that they leave out the actual theoretical part of the term—the "ontological". Lynch's engagement with the ontological, reworked as "ontography" (2013) and subsequently adopted partly by Woolgar and Lezaun (2015), point to more crucial issues in the discussion regarding this term, some of which I touched upon earlier (i.e., whether ontology is used to describe reality or to antagonise versions that understand it as singular).

Moving on to the second part of the question, the ontological one, the way it has been presented in STS, at least by Mol (1999), has obvious affinities with the new wave of anthropology, also termed an ontological turn. However, there is a fundamental difference between the ontological turn in STS and the one in anthropology. In the work of Viveiros de Castro (1998), Holbraand (2009), Descola (2013), for example, the attention to radical alterity is stipulated by the methodological need to do justice to conceptual differences in the point of view of indigenous peoples, without reducing it to colonialist schemes. The attention to ontologies there takes a multinaturalist note, as anthropologists adopt a more or less a priori stance to radical difference, thus the framework of ontological multiplicity risks ending up being just another traditional form of ontology. Although, as I mentioned before, Mol (1999)—paradigmatically—oscillates towards such a stance, the STS attention to ontologies becomes a matter of ontogenesis—i.e., it is less a methodological a priori but rather a way to meet the ontogenetic qualities of technoscience on the level of practice without reifying the said qualities. In other words, ontological inquiry in STS becomes a way to eventualise reality without, however, essentialising it.

Another disciplinary interlocutor to the ontological turn seems to be philosophy. Here, however, the picture is different. While anthropological inquiry into ontologies is in dialogue with STS[7] and is understood mostly as an ally, philosophy assumes the role of the foe. My hypothesis for the reason behind this animosity is that the word "ontology" has been encountered by STS researchers within the discourse of positivist philosophers of science, where the term is taken at face value. As a result, from an STS perspective, philosophy becomes the face of how *not* to perform ontological inquiries. Although this

[7] For example, in discipline-defining conferences such as the Americal Anthropological Association's annual conference in 2013.

stance is warranted for a large part of the philosophy of science, a possible ally from the side of philosophy could be some versions of contemporary critical theory, since in this tradition an ontological inquiry is almost always a critique of ontology (e.g., Foucault, 1997). Examples from the critical theory of social movements, where onto-poietic activity is at the centre of ontological inquiry, could be an overlooked ally in ontological STS inquiries (e.g., Rancière, 2004)[8].

[8] Without going into much philosophical detail, I would argue that critical theory enables us to understand the crafting of biomatter in the DBTL along the lines of bioengineering objects *as events* or, rather, as enactments of particular realities. This view would be in opposition to an essentialising paradigm that that would claim that bioengineering is manipulation, thus assuming the manipulation of something already pre-existing, a substratum preceding human intervention (such a view could re-introduce Nature and a quest for universal objectivity through the backdoor). This way, bioengineering could be better understood as a series of events, a series of onto-poietic activity: although the various objects of technoscience in the DBTL cycle are composed of previous parts of other organisms and their DNA, they are ontologically new, i.e., their emergence cannot be reduced to their parts or preceding ones. That is, simply because they have come into existence, their enactment itself performs and reinforces a certain vision of reality.

4 Research question

The overarching goal of my research is to understand the journey of biomatter in the bioengineering workflow of a Design-Build-Test-Learn cycle. Phrased according to the language of the theoretical framework I employ, the following question is the guiding one for my research:

- RQ1: "How is synthetic biological matter enacted throughout the bioengineering Design-Build-Test-Learn framework?"

This question can be further divided into three sub-questions:

- RQ1.1: "What kind of technoscientific entities are enacted in each phase of a Design-Build-Test-Learn cycle through processes of inscription?"
- RQ1.2: "How are the technoscientific entities of one phase within the Design-Build-Test-Learn cycle translated to the next?"
- RQ1.3: "What kind of script does synthetic biomatter acquire through the onto-poietic activity taking place within a Design-Build-Test-Learn cycle?"

My research objective is to show how biomatter is brought to existence through a chain of ontic transformations that happen throughout a DBTL cycle between various technoscientific entities. Thus, I aim to elucidate the ontogenetic process of bringing to existence bio-objects that perform social scripts and presuppositions in the process of their creation. In particular, I hope to demonstrate that the process of enacting bio-entities through a DBTL cycle consists of an onto-poietic activity where bioengineering decisions remain unexamined yet, at the same time, their default performance scripts biomatter according to a model of digital, binary logic.

5 Methodological approach

5.1 Overview

The focus of my analysis is the bioengineering workflow "Design-Build-Test-Learn" (DBTL), which is carried out in biology laboratories with advanced technological capabilities called "biofoundries" (Holowko, Frow, Reid, Rourke, and Vickers, 2020). Also phrased as "pipeline", "framework" or "cycle", the DBTL is essentially an application of synthetic biology in the context of metabolic engineering (Voigt, 2012), meaning that methods and tools from the field of synthesising artificial DNA are employed to optimise the metabolism of a given single-cell organism for industrial and market-oriented purposes. In the case of the DBTL, the ultimate goal of the experiment is to increase the production of a pharmaceutical or industrial substance of interest (for example, flavonoids) through the construction of novel artificial biological systems that conform to particular design criteria.

As its name indicates, DBTL is structured in four separate phases. However, I will approach it through a threefold structure, which I consider more appropriate for distinguishing the key moments in the journey of biomatter within it, borrowed by Lionelli (2014). In analysing the various contexts that big data appear in biological contexts, she used the terms "data journey" and "data travel" to denote "how biological data […] travel across research contexts, and the significant conceptual and material scaffolding used by researchers to achieve this" (Lionelli, 2014, p. 3). There, she identifies three stages of data travel: De-contextualisation (i.e., the primary formatting of data to become compatible with dataset criteria), re-contextualisation (i.e., the adoption of data in new research contexts), and re-use (i.e., the use of data to support new scientific breakthroughs).

Although methodologically I adopt Lionelli's approach to focus on a "journey" in biological settings as well as her threefold structure, I employ both differently. Contrary to Lionelli, the journey I present here, the journey of biomatter within a DBTL cycle, is framed in ontological terms. That is, as I have already showed in the "Theoretical framework" section, my focus is placed neither on databases nor on big data but on the ontic transmutations that synthetic biomatter undertakes throughout the various phases of a DBTL workflow. Thus, regarding the journey aspect, I modify the content of the idea so that it fits to the particularities of my primary material.

Similarly, the threefold structure (of de-contextualisation, re-contextualisation, and re-use) is employed in my research to snapshot moments in the DBTL cycle where biomatter can be seen in its different ontic instantiations. Thus, the content of the threefold structure is tailored, once again, to the particularities of the DBTL workflow and does not follow Lionelli's analysis. To that end, I will start my analysis at the moment that matter has already assumed its full biological form, tracing its transformation from the Design and Build phases to the Test phase; this will be the de-contextualisation snapshot. Then, I trace the transformation of biomatter from the Test to the Learn phase; this is re-contextualisation. I conclude at the moment that synthetic biomatter passes from Learn to a new Build phase (and from one DBTL cycle to the next), when the lessons learnt are supposed to lead in optimising the single-cell construct; this is re-use. The following table summarises the match between DBTL phases and my threefold structure.

Table 2: Match between DBTL phases and Lionelli's analytical structure

DBTL phase(s) and cycles involved	**Threefold structure used**
From Design 1 and Build 1 to Test 1	De-contextualisation
From Test 1 to Learn 1	Re-contextualisation
From Learn 1 to Design 2 and Build 2	Re-use

To carry out the research, I used two types of published scientific articles as primary sources. The first type of articles are cases that describe the step-by-step deployment of a DBTL cycle, essentially documenting the experiment after it has already been conducted. There, I focus on the "Methods" part of the texts, because the technical description of how a DBTL cycle is performed is located there. These materials will be analysed by identifying the statements that describe the ontic enactments and transformations of biomatter throughout a given DBTL cycle. In particular, verbs in those statements will be considered as markers of activity, i.e., indicators of where enactments of biomatter can be singled out and demarcated.

The second type of articles used as primary sources, complementing the first, document the design specifications according to which single-cell organisms are constructed in the Design phase of a DBTL cycle. I use these articles to shed light on a particular moment in the DBTL workflow (the "re-use" step in my analytical structure), as

practical cases of DBTL remain mostly silent on how Learn results feed into the design criteria of a new DBTL cycle (Design and Build phases). In this second type of articles, which I use to strengthen my argument, I will analyse pictorial representations of how DNA is assembled in artificially-constructed biological systems. These representations will be analysed by focusing on the schematic depictions of the process, describing their affinity with schematic depictions of electronic circuits in computer engineering.

5.2 Reasons for choosing published texts as primary sources

There are three reasons for choosing published scientific texts as primary sources. The first has to do with temporal and spatial constraints. A DBTL cycle might take several months with up to a year to conclude, therefore it would be impossible to study it in any ethnographic manner—for example, within the timeframe available for writing my thesis (approximately six months). In addition, there are currently no public biofoundries registered in Greece according to the Global Biofoundries Alliance registry (Hillson et al., 2019). These barriers made the ethnographic study of DBTL inaccessible to the resources I had available, without even factoring in accessibility restrictions to laboratories due to the COVID-19 outbreak. It would require many additional resources to "follow scientists through society" in a literal way, per Latour's famous phrase. Therefore, an ethnographic study of the DBTL workflow was not possible under the circumstances.

The second reason that led me to use published scientific articles as primary material has been dictated by the topic itself. Although DBTL is presented in the literature as a cohesive one-stop workflow, the reality is that its various stages are spatially and temporally dispersed, and some steps in biofoundries might not have been (yet) fully automated in-house. For example, in Carbonell et al. (2018, p. 8), the preparation of DNA parts was outsourced to commercial vendors in Germany and the USA. Similarly, the Agile Biofoundry, the largest public biofoundry in the USA and behind two of the articles I discuss in my analysis—Opgenorth et al. (2019) and Pomraning et al. (2021)—is significantly dispersed geographically, with locations all over the United States. As a result, published scientific articles reflect a privileged analytical viewpoint since they become the loci where the various stages of the workflow are pieced together and DBTL is presented in its entirety.

I could have conducted interviews with scientists involved in conducting DBTL experiments and used them as primary material. However, there is a practical reason for not doing so. There is a considerable number of scientists involved in carrying out a DBTL cycle, usually more than fifteen (judging from the average authors in the published articles I discuss). If I would pursue this, I would be limited to one laboratory, risking generalising from the particular application of DBTL in that specific laboratory, which would be difficult to do considering my research question.

The third reason for choosing to analyse published scientific articles has to do with the problems that arise from using first-person accounts as primary material. Following Hannah Landecker's argument in *Culturing Life* (2007), I am critical of a widely held assumption in STS, according to which first-person accounts (e.g., ethnographic studies, interviews) provide a more privileged access to scientific practice than published papers. Supporting this assumption, Landecker (2007) argues, is the belief that these accounts are "more authentic—or at least more recognizable as research" since they "have accessed the experience of the person" and thus "[p]ublished papers are, as they say, secondary sources" (p. 21). However, if researchers try to "describe things that are complex, diffuse and messy" (Law, 2004, p. 2), then it seems that even first-person accounts may not offer a privileged way to the web of meaning in technoscience (as it is often thought to be; see Mol, 2002).

5.3 Collection of primary sources

To collect my primary sources, I firstly searched the PubMed database with keywords such as "Design-Build-Test-Learn workflow/pipeline/framework", "genetic circuit design", "synthetic biology", "metabolic engineering", "AI biology" and a variety of combinations between them. As the initial results were not very hopeful, I identified scientific journals that appeared frequently in the search: *ACS Synthetic Biology*, *Oxford Synthetic Biology*, and *Nature Communications*, and scanned their back catalogue for full applications of the DBTL workflow. As this search, still, did not yield significantly more results, I started scanning review articles on DBTL as well as the ones I had already identified for relevant references (snowballing technique).

Following this process, over sixty scientific articles on various aspects of the DBTL workflow were identified and taken into consideration during primary data collection.

However, only a fraction of those described the application of a full DBTL workflow. In particular, twelve (12) were identified to explicitly employ a full DBTL framework (time range: 2018–2021). Thus, the rest of the articles were used to gain a general understanding of the workflow and its components, as it is interdisciplinary and involves expertise from multiple areas of synthetic biology and computer engineering. The following table presents the total scientific articles on DBTL identified during data collection in chronological order.

Table 3: Scientific articles identified that applied a full DBTL workflow

#	**Reference**	**Journal**
1)	Ji et al., 2022	*Metabolic Engineering*
2)	Moore et al., 2021	*Microbial Cell Factories*
3)	Pomraning et al., 2021	*Frontiers in Bioengineering and Biotechnology*
4)	Geiselman et al., 2020	*Microbial Cell Factories*
5)	Robinson et al., 2020	*Metabolic Engineering*
6)	Zhang et al., 2020	*Nature Communications*
7)	Feith et al., 2020	*Metabolites*
8)	Dunstan et al., 2020	*Synthetic Biology*
9)	HamediRad et al., 2019	*Nature Communications*
10)	Liu et al., 2019	*Biotechnology for Biofuels*
11)	Opgenorth et al., 2019	*ACS Synthetic Biology*
12)	Carbonell et al., 2018	*Communications Biology*

To highlight the "re-use" step in my analytical structure (i.e., the design specifications according to which single-cell organisms are constructed in the Design phase of a DBTL cycle), I decided to supplement the primary sources above with an analysis of pictorial representations that showcase my argument. To that end, I selected four pictures depicting genetic circuit design[9] from three review and research articles:

[9] In synthetic biology's jargon, this means the rational engineering principles according to which the metabolic system has been constructed in order to carry out a certain function (the language and the concept are both inspired by computer engineering).

Table 4: Scientific articles from which genetic circuit designs were taken from

#	**Reference**	**Journal**
13)	Brophy and Voigt, 2014	*Nature Methods*
14)	Becskei, Seraphin, and Serrano, 2001	*EMBO*
15)	Gardner, Cantor, and Collins, 2000	*Nature*

5.4 Challenges encountered during the primary data collection

The collection of primary sources proved challenging. The first reason was the scarcity of primary sources due to the emerging character of the DBTL framework. Initially, the framework was brought to the public eye by Amyris, a biotechnology company in California, that employed and standardised multiple cycles of DBTL (Nielsen and Kiesling 2016) to successfully manipulate the metabolic network of baker's yeast to produce artemisinic acid, a precursor of the drug artemisinin that is quite potent against the parasite that causes malaria (Paddon et al., 2013). However, it took Amyris about six to eight years to scale up and standardise this process (Radivojević, Costello, Workman, and Martin, 2020), which took off as a framework only after 2018. For example, prior to 2018 there are only nine total mentions of the DBTL approach in the PubMed database (two each year between 2015 to 2016; three in 2017); there is no mention before 2014, and its most popular year to date has been 2021 with 36 mentions. As the terms "synthetic biology" and "metabolic engineering" yield 5,217 and 6,911 mentions respectively in 2021 in the PubMed database, it is evident that the DBTL workflow is still in a nascent form. Correspondingly, the Global Biofoundries Alliance, a networking organisation that puts the dissemination of the DBTL workflow as one of its primary goals (Hillson et al., 2019), was inaugurated just before the COVID-19 pandemic hit, in November 2019.

The second reason for the scarcity of relevant primary sources is the capital and labour resources required to realise multiple DBTL cycles, which might make it accessible as a workflow only to a few and well-funded laboratories. For example, with regards to technology investment, only one piece of equipment that is considered standard in the workflow—a Hamilton liquid handler—may cost north of twenty thousand US dollars (Holowko, Frow, Reid, Rourke, and Vickers, 2020), without factoring in ancillary

equipment and the highly trained, interdisciplinary personnel required (biologists, computer scientists, coordinators, and so on).

Indeed, most of the articles documenting a full DBTL workflow come from a few national and high-profile private laboratories (DOE Agile BioFoundry and Lawrence Berkeley National Laboratory in the USA, SYNBIOCHEM and Earlham Institute in the UK). Larger and well-funded laboratories can handle the large datasets created by the DBTL workflow and invest resources for their curation (Lionelli, 2014), while smaller laboratories may specialise only in some aspects of the DBTL workflow, not necessarily needing to perform all the DBTL functions in-house. In addition, the implementation of one DBTL phase, at least in the early rollout of the workflow, may take several months (Qin et al., 2015), an investment in time that may be prohibitive for other laboratories.

The third reason for the scarcity of primary sources is the nature of my inquiry. Since my research required scientific articles that documented a full DBTL workflow, I could not include the largest part of the literature on DBTL as primary sources—namely, review articles or piecemeal descriptions of the DBTL workflow (i.e., dealing with only one aspect of the DBTL). Surprisingly, it was challenging to retrieve relevant material that included the last phase of the workflow, Learn. In fact, while collecting the primary sources it was evident that the Learn phase is the least developed of the four, thus researchers tend to under-report it (Whitford, Cruz-Morales, Keasling, and Weber, 2021).

5.5 Analysing primary sources

"The difference between a regular text in prose and a technical document is the stratification of the latter", writes Latour (1987, p. 48). Stratification, he continues, is the "surest sign that a text has become scientific", a "maze" where each claim "is interrupted by references outside the texts or inside the texts to other parts, to figures, to columns, tables, legends, graphs", which can again "send you back to other parts of the same texts or to more outside references".

The way Latour describes Guillemin's paper on growth hormone-releasing hormone (GHRH) is quite close to the experience of reading a paper about the DBTL framework. The entry point to the maze of a DBTL pipeline, in this case, is not going to be a

Latourian reader infused with Cartesian-like doubt. Instead, the protagonist is going to be biomatter, and more precisely the journey that biomatter takes throughout the DBTL bioengineering cycle. Therefore, here I will not be concerned with the modality of scientific statements as Latour (1987, pp. 22–23). If Latour's move is one of suspicion and doubt, in Ricœur's (1965/1972) terminology—where suspicion reveals more and more layers hidden behind statements—then the move I perform in conducting textual analysis aspires to be agnostic towards modalities and closer to that performed by a humble hound.

In the same way that hounds are trained to trace scents that they have never encountered before and are indifferent to their positive or negative modality, I adopt a similar approach to scientific statements. I do not have any prior training in biology, engineering, or computer science. Instead, as I, in my ignorance, trace the journey of biomatter across the DBTL workflow, I will take scientific statements as markers of activity, in a similar way that hounds perceive odours (Horowitz, 2009, pp. 34-67). I do not fully understand what those statements mean in the technical context of bioengineering (e.g., did the experiment go well or not), in the same way that a dog does not understand the context of an odour from a human perspective.

Approaching discourse in this light, statements-as-markers-of-activity indicate both a presence and an absence (and especially verbs in statements—as verbs are the linguistic markers that demonstrate action, occurrence, or state of being). That is, statements both describe a certain activity and stand to convey it (presence), yet they demand a cohort of intertextual references to be understood by a non-scientist (absence). This oscillation between the primary sources collected and the wider scientific references reviewed was unavoidable in my analysis.

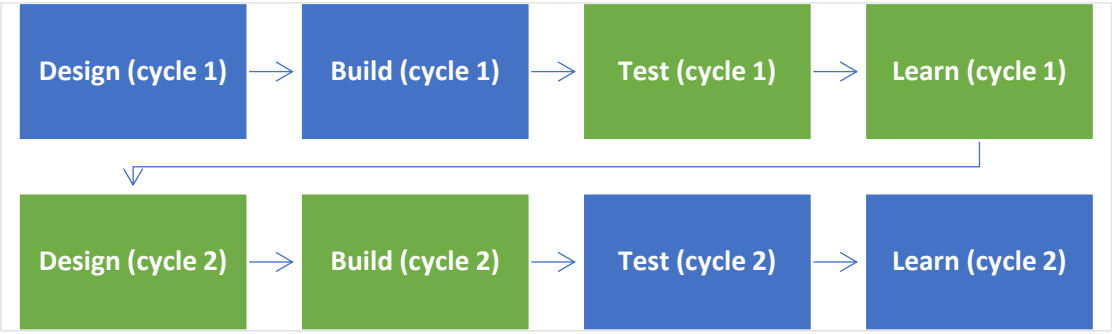
6 Analysis

6.1 Step 1: Decontextualization

To understand the journey of biomatter in the DBTL cycle, it is best to begin the analysis right after the initial Design and Build phases have been completed and right before the Test phase begins. There are two reasons for this. First, it is at that moment, after the first iteration of the Design and Build phases, that matter assumes its biological form: The single-cell artificial organism has been built and has not yet been taken apart through analytical chemistry screenings in the Test phase. All the initial design choices have been made during the Design phase (i.e., what will be the target molecule, what regulatory biosystem is most suitable to produce it, which host is the most compatible) through mathematical models of metabolism and BioCAD software, and the organism has been built. This is the moment of ignorance—the moment in which bioengineers do not yet know if their construct works.

Secondly, the first Design and Build iterations could not have yet incorporated the lessons learnt from the Test and Learn phases; only the subsequent Design and Build iterations do. Thus, starting the analysis during the passage to the first Test and Learn phases and moving on to subsequent Design and Built phases, I will be able to trace biomatter in a full DBTL cycle taking into account, crucially, how screening results feed into design choices in the workflow. The following figure depicts the start and end of my analysis (in green):

Figure 3: The focus of analysis



Entering the Test phase, biomatter undergoes certain high-throughput screenings. The objective is to acquire data on whether the transcription of the genome already being built has produced the desired chemical results at an industrially exploitable scale. This

is the first snapshot that I will describe in the analysis, the moment that a process of inscription leads to a transmutation in the ontic status of the synthetic biomatter.

Biomatter enters the Test phase as culture growth (DNA, proteins, peptides, membranes, and so on); essentially in a haptic, *wet biology* ontic status. However, at the end of the Test phase, biomatter assumes a radically different ontic status, that of an army of numerical traces in electronic form – data. During the heavy manipulation that biomatter-as-culture-growth undergoes during Test phase screenings, the single-cell organism is decontextualised (deterritorialised would be also appropriate) through a process of inscription (Latour, 1989), and from wet biology biomatter it ends up as data. This transmutation, it is worth noting, comes along with a shift in the infrastructural placeholder of biomatter (thus I find apt to name this step decontextualization), as there is an ontic shift from the environment in which biomatter finds itself in: The laboratory bench (or the robotic liquid handler) of wet biology (e.g., culture growths, mixtures, and so on) is substituted by the environment of an electronic computer.

This transmutation of biomatter from 'wet' to 'dry', from peptides to zeros and ones, and from biological to electronic traces, takes place in the following way:

> Peptides digests were diluted to 0.1 mg/mL with nanopure water for LC-MS/MS analysis. Five mL of samples were loaded onto inhouse packed reversed-phase capillary columns (70 cm × 75 mm i.d.) with 3 mm Jupiter C18 particles. The separation was carried out using a nanoAcquity HPLC system (Waters Corporation, Milford, MA, United States) at room temperature. The mobile phase A is 0.1% formic acid in water while the mobile phase B is 0.1% formic acid in acetonitrile. The elution was carried out at 300 nL/min with the following gradient: 0–2 min 1% B; 2– 20 min 8% B; 20–75 min 12%B; 75–97 min 30%B; 97–100 min 45%; 100–105 95%; 105–110 min 95%; 110–140 min 1%. The eluting peptides were directly analyzed using a Q Exactive HF mass spectrometer (Thermo Fisher Scientific) in data-dependent acquisition mode. Mass spectrometer settings were as following: full MS (AGC, 3 × 106; resolution,

> 60,000; m/z range, 300–1800; maximum ion time, 20 ms); MS/MS (AGC, 1 × 105; resolution, 15000; m/z range, 200–2000; maximum ion time, 100 ms; TopN, 12; isolation width, 2 Da; dynamic exclusion, 30.0 s; collision energy, NCE 30). Pomraning et al. (2021, p. 4).

The statements above, and in particular the verbs of the passage, become indicators of a long series of manipulations that result in the transmutation of biomatter from the culture growth to data: Peptides (biomatter) are diluted in a mixture; the mixture is loaded into the columns of a machine (nanoAcquity HPLC system) that separates them by measuring their interaction with another material (3 mm Jupiter C18 particles); biomatter acquires mobile phases and has its composition altered (elution with a gradient); the already heavily manipulated peptides enter another machine (Q Exactive HF mass spectrometer) which, by ionising them and then accelerating them through a small tube, will produce certain molecular traces (resolution, mass-to-charge range, ion time, and so on)[10].

This transmutation of biomatter takes place in almost identical ways in all primary sources. The method used for this transmutation of biomatter is an analytical chemistry method called liquid chromatography–mass spectrometry (LC–MS). In fact, as the DBTL workflow is customised to fit the experimental parameters and available resources of each case, it was observed that all primary sources used a slight variation of the said LC–MS method. For instance, in the example above, the method used is high pressure liquid chromatography (HPLC) and tandem mass spectrometry (MS/MS). The principles behind each variation across other primary sources remains the same, with the difference being the quantification precision and the complexity[11].

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[10] An example of how such data look like, taken from Pomraning et al. (2021), can be found in https://www.frontiersin.org/articles/10.3389/fbioe.2021.603832/full#supplementary-material

[11] The only outlier in this has been HamediRad et al. (2019), where the authors develop an integrated robotic system coupled with machine learning algorithms, BioAutomata, with the aim to fully automate the DBTL cycle. However, as they note:

> Potential challenges of a universal application of BioAutomata for pathway optimization include extraction methods that are difficult to perform on an automated platform, or analytical/quantification methods that require equipment more complex than a plate reader, such as Gas Chromatography-Mass

Briefly put, this method can separate mixtures (the LC part) and provide information regarding their structural molecular identity with high sensitivity (the MS part). MS is especially crucial for the identification and quantification of molecules, as it can provide information, such as molecular weight and volume, which are essential for the identification of their structural and chemical properties. MS works by ionising the sample and accelerating it through a small, curved vacuum tube. As the ionised molecules pass through the tube, they take slightly different routes, due to mass difference. At the end of the tube there is a detector, where the ionised molecules collide. This collision imprints information regarding their speed, time, weight, and volume. This information are then compared with known values of molecules, usually held in protein and peptide libraries, yielding information on which molecules were mostly present in the sample and in which volumes. Through this method, therefore, the target compound, as well as the peptides and proteins that led to it, can be identified and quantified.

Let us see another example:

> Peptides were analyzed using an Agilent 1290 liquid chromatography system coupled to an Agilent 6460 QQQ mass spectrometer (Agilent Technologies, Santa Clara, CA). The peptide samples (20 μg loaded on column) were separated on an Ascentis Express Peptide ES-C18 column (2.7 μm particle size, 160 Å pore size, 5 cm length × 2.1 mm i.d., coupled to a 5 mm × 2.1 mm i.d. guard column with similar particle and pore size; Sigma-Aldrich, St. Louis, MO), with the system operating at a flow rate of 0.400 mL/min and the column compartment at 60 °C. Peptides were eluted into the mass spectrometer via a gradient with an initial starting condition of 95% Buffer A (99.9% water, 0.1% formic acid) and 5% Buffer B (99.9%

> Spectrometry (GC-MS) or Liquid Chromatography-Mass Spectrometry (LC-MS) instruments. (HamediRad et al., 2019, p. 6).

For this reason, their choice of biosystem optimization, the lycopene biosynthetic pathway, was chosen specifically due to its easier identification and quantification method ("Lycopene can be extracted by organic solvents and quantified calorimetrically by measuring the absorbance"; HamediRad et al., 2019, p. 8).

> acetonitrile, 0.1% formic acid). Buffer B was held at 5% for 0.2 min, then increased to 35% B over 5.5 min. Buffer B was further increased to 80% of 0.3 min, where it was held for 2 min, then ramped back down to 5% B over 0.5 min, where it was held for 1.5 min to re-equilibrate the column to the initial starting condition. (Opgenorth et al., 2019, p. 1347)

Here, as in the previous quote, the transmutation happens through the same analytical chemistry process, LC–MS ("Agilent 1290 liquid chromatography system coupled to an Agilent 6460 QQQ mass spectrometer", Opgenorth et al., 2019, p. 1347). In the beginning, biomatter enters the Test phase as in its haptic ontic status (peptides). Then, the same steps are set in motion, with slight variations only in details (e.g., the size of the column used for peptide separation): Peptides are separated by measuring their interaction with another material (in the Ascentis Express Peptide ES-C18 column); the composition of the peptides is altered by gradient elution, with peptides acquiring mobile phases. Here, however, the description omits the part where peptides are ionised and then accelerated in the mass spectrometer, as this step is already presupposed by mentioning the "Agilent 6460 QQQ mass spectrometer" in the beginning. I will not quote from any other articles, since the process is essentially the same with slight variations.

By travelling from the Build to the Test phase, biomatter is enacted as two different ontic states: Both as a biological trace and an electronic one. The electronic state is supposed to mirror the biological and stands in (quite literally) for the biological trace (in the passages above, peptides), becoming its double and summoned to represent it (Haggerty and Ericson, 2000). Through this inscription process, the ontic status of the biological matter is double, split between two equally existing ontologies; the molecules, the peptides, and so on still exist, but now another ontology has emerged, with a radically different ontic status, which is forced to take its place.

The process of inscription that takes place by passing from the Build to the Test phase is simultaneously the moment that the biological trace is deterritorialised, the moment that a new technoscientific object emerges in the DBTL workflow, and the moment that two different ontic states—the biological and the electronic trace—are summoned to

establish a link of inter-intelligibility. Here we can recall, as Mol argued, "objects are framed as parts of events that occur and plays that are staged" (2002, p. 44). The triple moment of deterritorialization, emergence, and translation is the "event" in Mol's terminology, and the "plays that are staged" are the inscription processes and devices involved in passing from the Build to the Test phase (mainly the Liquid Chromatography–Mass Spectrometry machine and screening technique). The "objects" in question are the two traces that we are left with at the end of the Test phase: The biological and the electronic. The link established between the electronic computer and wet biology reveals biomatter as a double, Janus-faced, object with two distinct ontologies.

6.2 Step 2: Recontectualisation

At the end of the Test phase, we encounter two objects and two ontologies which biomatter has occupied in the DBTL workflow, the biological trace within the wet biology environment and the electronic trace within the electronic computer environment. Moving towards the Learn phase, the electronic trace will be enacted again as a distinct object, remaining however within the electronic computer environment. In this section, I will show the emergence of a third object, the visual trace, as the ontic status of biomatter will radically shift again. It is obvious that the enactment of biomatter in the Learn phase is ontologically the furthest from the original position of the biological trace (Design/Build).

The emergence of this third object is less standardised compared to the emergence of the previous object (electronic data). This is the point that primary sources start to efface the labour required to pass from the electronic to the visual trace. In fact, only four out of the twelve articles mention that there was extensive data clean-up involved after the data were obtained from the LC–MS process during the Test phase. Consider the following passage:

> All the LC-SRM data were imported into Skyline software and the peak boundaries were manually inspected to ensure correct peak assignment and peak boundaries. Peak detection and integration were determined based on two criteria: 1. The same LC retention time and 2. Approximately the same relative peak intensity ratios across

> multiple transitions between the light peptides and heavy peptide standards. The total peak area ratios of endogenous light peptides and their corresponding heavy isotope-labeled internal standards were then exported from Skyline software as Ratio-to-Standard. For each peptide, the total peak area ratios of individual samples were normalized to the average total peak area ratio of all the samples. For each sample, protein abundance was calculated as an average of the normalized total peak area ratios of all three peptides of a protein. (Geiselman, 2020, p. 10).

It is clear from the passage above that there is a lot of noise in the electronic traces produced during the Test phase, as also confirmed by the relevant synthetic biology literature (Mey, Clauwaert, Van Huffel, Waegeman, and De Mey, 2021). There needs to be a manual inspection of the data to ensure consistency, based on certain criteria; the peaks are normalised and protein abundance is calculated. Data clean-up descriptions appear in a similar way in other primary sources. However, apart from such mentions in Pomraning et al. (2021) and Opgenorth et al. (2019), all other primary sources remain silent on this matter. This seems to denote a textual lapse into opaqueness, as the labour and the human choices involved in the curation of data are not explicitly discussed.[12]

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[12] In fact, in Carbonell et al. (2018) there is a dedicated section describing "Data management support", providing a glimpse of the vast array of knowledge and infrastructure required to render the data acquired meaningful:

> The integrated pipeline benefited from the support of a data management system consisting of commercial, open source, and bespoke data management platforms, which assisted in making the data FAIR (findable, accessible, interoperable, reusable) throughout the pipeline. An instance of the open-source JBEI Inventory of Composable Elements (ICE) registry was used as registry of parts, plasmids, and strains. Recording of experiments was performed using shared electronic lab notebooks (http://www.labarchives.com). A data acquisition service was developed in house, which allowed data to be remotely transferred from laboratory instruments (e.g., QqQ), archived and backed up in our large data store (Synology NAS Disk Station with mirrored backup). During transfer, data were simultaneously ingested into our OpenBis software along with associated metadata for easy retrieval. (Carbonell et al., 2018, p. 9).

Multiple data management platforms, a registry, an online documentation space, a data acquisition service, data storage facilities, and heavy use of bespoke and in-house software programmes—are all essential infrastructure in the DBTL, not to mention the capital and labour resources required to coordinate, maintain, curate, and operate it. The electronic trace of biomatter travels, this time according

The opaqueness becomes thicker, and the scarcity of information is more prevalent, when the electronic trace undergoes mathematical analysis to identify which DNA constructs were more successful in producing the required substances in optimum volume. Here, apart from the opaqueness, we encounter quite a fragmentation in the method this analysis is carried out in the primary sources. This is not surprising, however, as the synthetic biology literature considers this particular phase of the workflow the least standardised, as it is "nonsystematic and lacks statistical rigor, relying on ad hoc observations, literature data, and intuition gathered by individual researchers responsible for the next round of pathway design" (Nielsen and Keasling, 2016, p. 1193).

Nevertheless, the mathematical analysis in the Learn phase of the DBTL is crucial for the transmutation of the electronic trace into a visual one. It is this analysis that—whether based on statistical or on algorithmic methods of inferring correlation (e.g., machine learning)—ingrains order among the electronic traces. This ordering of electronic traces happens in such a way that it can then be visualised: A new object emerges in the DBTL workflow. The mathematical analysis aims to identify correlations between, on the one hand, DNA builds (and therefore peptides and proteins) and, on the other hand, the concentration of target molecules so as to identify the DNA builds that were successful and could be further optimized to yield higher titers of the target molecules.

Despite the lack of standardisation, this thesis's primary sources can be categorised into three groups on this issue according to the way the mathematical analysis of the electronic trace takes place: Articles that do not mention the method of mathematical analysis (Ji et al., 2022; Moore et al., 2021; Gieselman et al., 2020; Liu et al., 2019), articles that carry out standard statistical analysis (Pomraning et al., 2021; Dunstan et al., 2020; Feith et al., 2020; Robinson et al., 2020; Carbonell et al., 2018), and articles that carry out the analysis through algorithms—machine learning, more precisely (Zhang et al., 2020; HamediRad et al., 2019; Opgenorth et al., 2019).

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to Lionelli's (2014) original conception, between all these various settings and contexts while at the same time the work required to carry it out is edited out.

Regarding the first group, it is surprising that the method of mathematical analysis is not mentioned. Yet this corroborates the view in the synthetic biology literature that the Learn phase is very often based on scientists' own knowledge, intuition, and ad hoc analysis (Nielsen and Keasling, 2016). This omission also confirms the observation that the further we follow the journey of biomatter within the DBTL workflow, the more we encounter an effacement of much of the labour and tacit knowledge required. Regarding the other two groups, there is a clear geographical aspect to it. Standard statistics are deployed mostly by researchers in Europe (the UK and Germany, and the SYNBIOCHEM biofoundry of the University of Manchester, in particular; the only exception being Pomraning et al., 2021), and the machine learning approach is deployed by researchers in the USA (specifically, in the Agile BioFoundry, which is funded by the U.S. Department of Energy).

The activity of mathematical analysis performed on the electronic trace is described briefly in all the articles belonging to the second group, presumably due to the well-known nature of the statistical methods which are not native to biology, as the following passage shows:

> Statistics were performed with established standard methods (Webb-Robertson et al., 2017). For time we utilized an ANOVA with a Dunnett's test to compare all time points back to the first time point within each strain. We also utilized a g-test with a Bonferonni correction to identify qualitative markers both to compare strains or time (Webb-Robertson et al., 2010). (Pomraning et al., 2021, p. 4)

Like other articles of this group, the statistical analysis is described in the following way:

> The linear model for standard curves was selected based on the analysis of data by linear regression weighting factors 1/x, using MassLynx V4.1 SCN905, with TargetLynx (Waters Corp., Milford, MA, USA). (Dunstan et al., 2020, p. 4)

In contrast, the description of the machine learning used during the Learn phase in the third group is described in length, presumably due to the novelty of the approach in

treating this kind of data. Here the relevant passages are quite long on the primary sources, with Opgenorth et al. (2019) being the only to provide detailed mathematical formulas, while Zhang et al. (2020) is based on already fully-fledged machine learning approaches, ART and EVOLVE, developed by other researchers in previous scientific studies (Radivojević, Costello, Workman, and Martin, 2019). Here I will cite from the second source regarding the use of the ART algorithm:

> In the ART approach, outliers were identified and removed based on replicate differences in GFP synthesis rate relative to the mean value for the strain. Replicates with the one percent most extreme differences were identified and the corresponding strains were removed. GFP synthesis rate was modeled as a function of promoter combination, represented through one-hot encoding, using the ART. Briefly, ART uses a probabilistic ensemble model consisting of eight individual models. The weight of each ensemble model is considered a random variable with a probability distribution characterized by the available training data, and determined through Bayesian inference and Markov Chain Monte Carlo. ART uses the trained ensemble model in combination with a Parallel Tempering approach to recommend 30 promoter combinations (unseen designs), which are predicted to improve production. The recommended designs were chosen as the 30 strains with the highest expected GFP synthesis rate predicted by the model. This recommendation approach was labeled exploitative since predictions with high uncertainty were not prioritized, although ART can provide both exploitative and explorative recommendations. (Zhang et al., 2020, p. 11)

Here, once again, we observe the activity of crafting that goes into shifting from the data object to the visual object within the ontology of the electronic computer; for example, outlier and replicate strain parameters need to be identified and removed. However, in this third group, we observe a very high level of automating the process through machine learning. The recommended design changes that ART produces, or any other algorithmic approach in the third group, is the opaquest point in the whole workflow.

Despite that, the outcome of all three approaches in the primary sources (no mention of the method, statistical approach, machine learning approach) leads to visual traces, "the 'window' in a technical paper" (Latour, 1987, p. 68)—pictures that depict the outcome of the long inscription process that started with the deterritorialization of biomatter in the Test phase (see the "Decontextualisation" section of this thesis). Here, the ontic status of the biological matter changes again. As data are cleaned-up and morphed into a structured hive through a statistical or algorithmic process, they end up becoming a visual trace, (usually in the form of a correlational graph or a heatmap). Such visual traces inform us that certain genes (synthetic DNA constructs) were better than others in transcribing themselves into RNA, then into peptides, and then into proteins, which then produced larger concentrations of the target molecule than other genes.

The recontextualization of the biomatter takes place here. After it shifted into a radically different ontological environment by passing from the Build to the Test phase and from the wet biology to the electronic computer, now it undergoes further curation within the ontological environment of the computer to end up in the Learn phase, where its ontic status is transmuted once again; from numerical data, biomatter morphs into a picture. Thus, the visual trace is another ontic transmutation of biomatter in its journey within the DBTL workflow.

6.3 Step 3: Reuse

At the end of the Learn phase, the ontic state of matter is the furthest from its biological form, from the beginning of its journey, which we started to follow in the Build phase during "Decontextualisation". Here, at "Reuse", the focus will be on the moment that the matter passes from the Learn phase to a new Design/Build one, thereby concluding the tracking of biomatter through a full DBTL cycle. In this transitive moment, biomatter shifts back from the digital ontology of the electronic computer to the ontology of the wet laboratory. It is at this step that the process of scripting biological matter and the design choices involved in it become apparent in the workflow.

So far, we have followed biomatter through three phases of the workflow and its corresponding bio-objects: From a biological state it was transmuted into an electronic one (data) during the Test phase, and from data it shifted into a visual form in the Learn phase (picture). Now, moving towards the Design phase, biomatter assumes yet another

form, being enacted as an architectural structure (DNA code), before finally turning into its biological form once again when passing from the Design to Build.

At this step, the "Reuse", I argue that we can identify the appearance of an ontological script that acts as a unifying equaliser between the various enactments of biomatter, materially inscribing the binary logic of an electronic circuit into the biosystem functions of a single-cell organism. To that end, in this section, I will show the trace of another enactment, that of biomatter being enacted as an architectural structure (of DNA), with the ontic status of biomatter radically shifting again. This is the final ontic form in the journey of biomatter in the DBTL workflow, and in all subsequent cycles the same enactments will be repeated over and over again.

The circular flow of the DBTL entails that the lessons learnt from the Test and the Learn phases will enrich the Design and Build phases of a subsequent workflow cycle. During previous steps, the processes leading to the corresponding enactments were to be found in the "Methods" parts of texts that describe full application cases of the DBTL workflow. However, the activity indicating the labour and the scientific choices involved in passing from the Learn to a new Design phase are entirely effaced from such texts. The only traces of activity are passages such as the following:

> Coding sequences for TcPAND, BcBAPAT, and EcHPDH were codon optimized for Aspergillus species and synthesized. (Pomraning et al., 2021, p. 3).

There is no description of the optimisation logic followed by the authors, as they go on to describe only the DNA pairs involved. This was also the case for all other articles, as none of them mention what kind of optimisation took place. To be sure, there are parts of the primary sources, outside of the "Methods" part, that discuss how the design of the pathway took place:

> First, we deleted the tyrR gene (a transcriptional regulator of aromatic amino acid biosynthesis) from the host cell genome, followed by deletion of either the pheA or tyrA genes (chorismate mutase/prephenate dehydratase (CMPDH) for phenylalanine and tyrosine biosynthesis, respectively). For pathways with aromatic

> aldehyde intermediates we also deleted the feaB gene (phenylacetaldehyde dehydrogenase). (Robinson et al., 2020, p. 173).

However, they never explain the design logic driving such deletions in the pathways. One is never treated with an explanation on why to substitute promoter X with a promoter Y, for example. The endpoint of design is never revealed, only the procedure undertaken to insert or delete DNA.

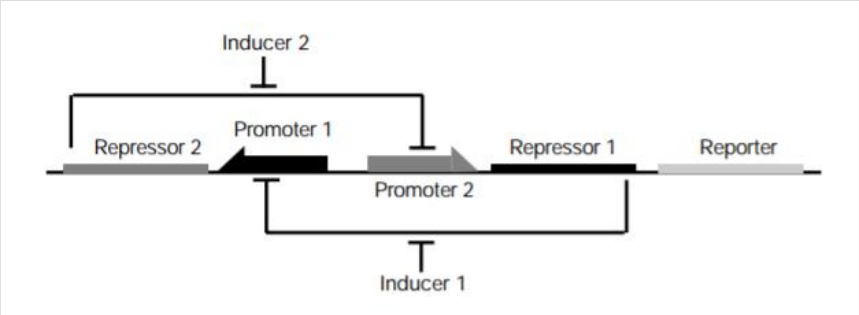
Across all primary sources, there are descriptions of how the new DNA builds were constructed, and there is already the well-understood assumption that optimisation in this context means to "fine-tune the expression of genes involved in [the] biosynthesis (the inputs to the function) to achieve the highest […] production (output of the function)" (HamediRad et al., 2019, p. 2) of a target substance. But what is omitted in such descriptions is the design principles of how exactly optimisation acquires meaning when optimising a 'genetic circuit', as it is called the design of a synthetic biosystem in the synthetic biologists' jargon. In other words, the design script of optimization in DBTL does not appear in the primary sources that document full applications of the workflow.

If we return to the hound analogy as presented in the "Methodological approach" section, the scent of biomatter is lost here. However, to avoid this dead end and continue to track the journey of biomatter in the DBTL workflow, we need to do what a trained hound would do: Take a step back from the current route and start looking for clues in the vicinity. In the vicinity of the primary sources documenting a DBTL workflow, not surprisingly, we encounter another area that the DBTL has 'visited', another marker of activity—this time the action is not in a verb form but codified as a sign. This area is genetic circuit design, i.e., the logic alongside which the biosystem of a synthetic cell is designed, where the markers of codified activity are the pictorial representations of the biosystem synthetic designs[13].

[13] For the full explanation of why this step is needed methodologically, please see the "Methodological approach" section.

Equipped with such new resources, we can continue to follow biomatter in the DBTL and zoom into what meaning optimisation acquires in this bioengineering context. Let us first look at one of the earliest and most well-known gene circuits, the toggle switch:

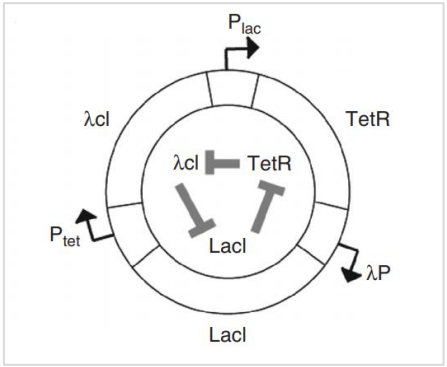
Figure 4: "Toggle switch design" as appears in Gardner, Cantor, and Collins, 2000, p. 339.



The name of the design, *toggle switch*, refers to its ability to switch between 'on' and 'off' states, thus the circuit can exhibit a bistable behaviour. It comprises two repressors (proteins that 'close' or 'open' the possibility to express a gene), which can mutually repress each other. The inducers are the inputs that flip the switch between the binary states of the circuit; for example, when inducer 1 is introduced to the biosystem, that input inhibits repressor 1, and repressor 2 will be highly expressed, and thus, the reporter will be 'off'. Conversely, when inducer 2 is added to inhibit repressor 2, repressor 1 will be highly expressed and the reporter will be 'on'.

Let us look at another example of a design of a genetic circuit, the repressilator[14].

Figure 5: "Repressilator gene circuit layout", as appears in Becskei, Seraphin, and Serrano, 2001; here adapted from Tu, Lee, Ozdere, Lee, and You, 2007, p. 364.



[14] Although the original design comes from Becskei, Seraphin, and Serrano (2001), here I will use a simplified design as reproduced in Tu, Lee, Ozdere, Lee, and You (2007, p. 364).

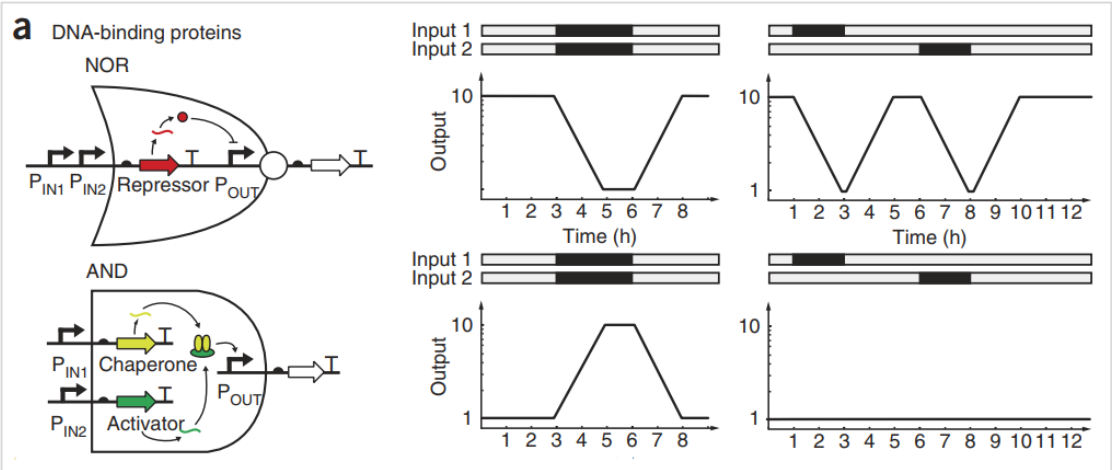
This picture shows three repressors sequentially repressing each other (TetR, lcI, and LacI). The first repressor (TetR) inhibits the second repressor (lcI), whose expression, in turn, inhibits the third repressor (LacI); now the third repressor (LacI) inhibits the first repressor (TetR). This design performs a negative feedback loop with a long cascade.

By analysing such designs, it becomes apparent what kind of pathway design the deletion and insertion of DNA mentioned above perform. Optimisation is a reduction of the analogical circuits in biosystems to binary signals in the same way that electronic ('digital') ones are designed in electronic circuit boards. The rhetoric and iconography of synthetic biology regarding the simplification of biological complexity comes full circle, spilling over metaphors and infiltrating in the logic that matter is designed.

The editorial of the *ACS Synthetic Biology* journal, discussed at the beginning of the "Introduction" section of this thesis, routinely framed DNA as having "idiosyncrasies". What synthetic biology aims to carry out through the DBTL workflow is to turn the 'analog', fuzzy and continuous signals of historical biosystems into discreet, binary signals by jamming together various parts of DNA to bring to life a synthetic single-cell organism that exemplifies such a 'digital' electronic engineering logic.

Now let us take a look at another type of a genetic circuit design in synthetic biology, the logic gates:

Figure 6: "Logic gates", as appears in Brophy and Voigt, 2014, p. 508.



The picture above shows two types of synthetic logic gates, NOR and AND types as well as, on the right, how the gates respond when the inputs are introduced at the same time (center) or sequentially (right). On the top left corner, the NOR gate represents a repressor that needs two types of input (P_{IN1} and P_{IN2}) to turn 'off' the output (P_{OUT}). On the bottom left corner, the AND gate is based on an activator (receiving P_{IN1}) that requires a second protein (chaperone) to be active (receiving P_{IN2}). Based on these types of logic gates, more complex ones can be constructed, with multiple inputs based on truth tables (Roquet and Lu, 2014).

Returning to the DBTL workflow, now we have the missing piece in the moment that biomatter passes from the Learn to the Design phase. The optimization is occurring based on a binary digital/electronic logic. The visual trace in the Learn phase has a guiding function, providing lessons on how to optimise the initial synthetic DNA in the biological enactment of biomatter (Build phase). The logic guiding this optimisation is that of electronic engineering; the deletions and insertions of DNA, guided by the visual ontic state of biomatter, aim to enact an architectural arrangement (DNA) according to the digital logic of the electronic computer.

Tracing the passing of biomatter from the Learn to the Design phase, the visual trace collapses back into new DNA code, bringing to life the final bio-object within the digital environment of the electronic computer. Then, by realising the optimised DNA architectural structure by passing from the Design to the Build phase, the architectural trace of biomatter is transmuted again into the biological one, and correspondingly biomatter shifts back from the electronic computer ontology to the wet biology one, completing an ontological oscillation alongside a full DBTL cycle.

7 Conclusion

In this section, I first summarise the results of my analysis. Then, I discuss the results in the context of the theoretical framework I employed, giving particular emphasis to the ontological politics involved in the Design-Build-Test-Learn synthetic biology workflow I researched in this thesis. I end on a note on artificiality, regarding potential lines of research that could be further explored.

7.1 Results

My research aimed to understand under what processes of inscription, translation, and scripting biological matter is enacted as a single-cell organism in the iterative synthetic biology workflow called "Design-Build-Test-Learn". Following closely the journey of biomatter in cases that documented this workflow, I identified that two distinct ontological environments participate in its crafting: The wet biology infrastructure and the digital infrastructure of the electronic computer. Within the DBTL workflow, biomatter travels back and forth between those two ontologies, and this oscillation between the two is what enacts biomatter as a multiplicity of traces within the workflow.

Analysing the process of inscription and translation when passing from the Build to the Test phase, I identified that the biological trace of biomatter (single-cell organism) is transmuted into an electronic one (data); when passing from the Test to the Learn phase, the electronic trace is transmuted into a visual one (picture); when passing from the Learn to the Design phase, the visual trace is transmuted into an architectural one (DNA constructs); and finally, when passing from the Design to the Build phase, the architectural is transmuted into a biological trace once again (single-cell organism). As seen in the Table below, I identified four distinct enactments of biomatter that are part of the DBTL workflow: The biological (wet biology environment), the electronic, the visual, and the architectural (electronic computer environment).

Table 5: Enactments of biomatter identified per DBTL phases and the corresponding ontological environments involved

DBTL phase	**Analytical step**	**Enactments of biomatter**	**Ontological environment**
Build to Test	Decontextualisation	Biological to electronic	Wet biology to electronic computer

Test to Learn	Recontextualisation	Electronic to visual	Electronic computer
Learn to Design	Reuse	Visual to architectural	Electronic computer
Design to Build		Architectural to biological	Electronic computer to wet biology

To properly describe the passing of biomatter from visual to architectural (i.e., the design specifications according to which single-cell organisms are constructed in the Design phase of a DBTL cycle), I analysed pictorial representations from the primary sources which conceptually support DBTL cases. By doing this, I argued that there is an ontological script that acts as a unifying equaliser between the various enactments of biomatter. This script materially inscribes the single-cell organism with the logic of an electronic circuit through the orchestrated ontological choreography of enactments taking place throughout the DBTL. That is, the logic of an electronic circuit materialised in the functions of a single-cell organism is the outcome of the processes of inscription, translation, and scripting occurring when the wet biology ontology and the digital infrastructure ontology of the electronic computer are summoned to communicate and become aligned within the DBTL workflow.

In this way, I was able to show that the enactments that take place in the DBTL contribute towards a common computer engineering script that is being passed on and inscribed in the synthetic metabolic pathways of the single-cell organism. Overall, I showed that the activity of crafting synthetic organisms in synthetic biology workflows consists of an onto-poietic activity where the mode of communication between infrastructures with distinct ontologies inscribes performance scripts to biomatter according to the logic of that communication.

7.2 Discussion

Having analysed the DBTL workflow, we can now understand the double meaning of decoupling genetic systems from "the idiosyncrasies of DNA", as mentioned in the first lines of my introduction. Decoupling, or reducing biological complexity, in the DBTL workflow occurs not only on the level of materiality (biomatter) but also on the level of thinking (problem-solving). The reduction of biological complexity is obtained by synthesising biosystems inscribed with the logic of an electronic circuit (material level). Thus, the logic of decoupling exemplified in the onto-poietic process of the DBTL

mirrors the binary, ‘electronic’ input–output logic exemplified in the problem-solving mode of thinking in synthetic biology. The reduction of a complex socio-technical situation (e.g., energy production) as a purely technological problem (e.g., fuel shortage) that requires its corresponding technological solution (e.g., production of synthetic fuel through synthetic biology) displays the same ‘electronic’ mode of thinking inscribed in the single-cell organism that comes out of the DBTL. In the same way that synthetic biomatter is enacted on an ontic, material level as a binary computor that calculates biochemical information in an input–output format, problems are framed and solutions are calculated according to a binary, input–output logic in synthetic biology.

For this reason, I consider my research to be showcasing in material terms what Rabinow has called biosociality, a concept discussed in detail in the “Theoretical framework” section. That is, my analysis enables a material understanding of how biosociality is performed, where the modelling of nature (engineering of single-cell organisms) is based on culture (the design script of DBTL). This, in turn, showcases the value of this thesis. Tracing the crafting of multiplicity in technoscientific settings is a way to eventualise—to institute dispute where there was none before—sites of production on the ontic, material level (and not only on an epistemic one), sites where artefacts come into being and intervene in reality by performing their ontological script.

As a result, my analysis addresses technoscience on the level of onto-poiesis, attending to the political economy of ontologies in technoscientific settings, i.e., the material production and circulation of bio-objects. Building on the conceptual threads I discussed in the “Theoretical framework” section regarding ontological politics, I consider this approach affiliated with the history of political economy broadly understood, because attending to the mechanism of ontic difference production in sociotechnical arrangements reveals the agonistic character of enactments in technoscientific settings, as shaped through processes of inscription, translation, and scripting. In the case of biomatter in the DBTL, it means that such an approach enables us to understand that its production and circulation in such a bioengineering workflow is not just an ontological choreography (Cussins, 1996) but political negotiations taking place in an ontic, material language—yet away from the public scrutiny that is akin to such negotiations.

This is also the value of employing an ontological framework, as it enables us to understand conceptual frameworks and research methodologies as both normative and onto-poietic. If synthetic biology intervenes in generating materially and descriptively the ontic fabric of reality, then STS-inclined research on synthetic biology workflows performs an equally interventionist gesture: It interferes on the level of the associations established between materials, objects, and their descriptions and challenges them, producing different accounts of intelligibility of what synthetic biology is and does.

In this research, I asked what kind of artificiality is enacted, what kind of matter is brought to existence in this latest, high-tech version of biotechnology. Instead of examining what artificiality would mean for the concept of life, which is a question with metaphysical undertones, I have opted to ask what kind of onto-poiesis is taking place, a question about the contingencies of political and social ontologies. The question about the distinction of nature and culture, so much debated in STS and the humanities, becomes irrelevant through the historical emergence of biotechnology. Arguably, the question should be what kind of hybridities are enacted. Cyborg ontologies are flourishing in synthetic biology, in a way that is both faithful to and complicates Donna Haraway's political myth.

In conclusion, I would like to point out some lines of inquiry that could be further explored in the context of my research. First, the journey of biomatter could be tracked beyond the DBTL, when the single-cell organism leaves the biofoundry and travels to industrial settings. By investigating the journey of biomatter in various contexts, for example in the biofuel or the medical industry, there could be connections drawn with the concept of big science in biology, which could, in turn, enable the elucidation of the nexus between national funding, industrial applications, and synthetic biology research. Secondly, the theoretical framework employed in this thesis could be fine-tuned and further elaborated by drawing from the historical studies of science and biology, as well as from the strands of contemporary philosophy that problematise the concept of ontology.

Thirdly, regarding the methodology of the thesis, it could be potentially fruitful to track the journey of biomatter by employing ethnographic approaches. In addition, in order to refine the analysis of verbs as markers of activity, it could be beneficial to draw from

discourse analysis approaches that focus on typologies of verbs. Finally, the modes of communication and negotiation that have been established between the biofoundry and policy rooms, two sites of knowledge production regarding our understanding of synthetic biology, could be further explored. In particular, it would be worthwhile to track the journey of biomatter into policy rooms and back to biofoundries, situating the DBTL workflow across a variety of sites of material production across the synthetic biology value chain.

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