



Exploring biological possibility through synthetic biology

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Abstract

This paper analyzes the notion of possibility in biology and demonstrates how synthetic biology can provide understanding on the modal dimension of biological systems. Among modal concepts, biological possibility has received surprisingly little explicit treatment in the philosophy of science. The aim of this paper is to argue for the importance of the notion of biological possibility by showing how it provides both a philosophically and biologically fruitful category as well as introducing a new practically grounded way for its assessment. More precisely, we argue that synthetic biology can provide tools to scientifically anchor reasoning about biological possibilities. Two prominent strategies for this are identified and analyzed: the *design* of functionally new-to-nature systems and the *redesign* of naturally occurring systems and their parts. These approaches allow synthetic biologists to explore systems that are not normally evolutionarily accessible and draw modal inferences that extend in scope beyond their token realizations. Subsequently, these results in synthetic biology can also be relevant for discussions on evolutionary contingency, providing new methods and insight to the study of various sources of unactualized possibilities in biology.

Keywords Biological possibility · Synthetic biology · Design · Modality · Evolutionary contingency · Biological constraints

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1 Introduction

This paper analyzes the notion of biological possibility and demonstrates how synthetic biology can provide understanding on the modal dimension of biological systems. The discussion of biological modal concepts has usually focused on the contrast between evolutionary contingency and convergence, and the subsequent question of how sensitive evolutionary outcomes are to changes in their evolutionary pathways (e.g., Beatty, 2006; Gould, 1989). However, with the exception of Dennett (1995), biological possibility per se has received relatively little explicit treatment in contemporary philosophy of biology. The main reason behind this underrepresentation of biological possibility lies in the supposed speculative nature of the concept. Not only have the prospects of a substantial notion of possibility been contested on philosophical grounds (e.g., Quine, 1953), there are many completely pragmatic reasons that make scientists wary of unnecessary commitments to purely hypothetical entities.

The aim of this paper is to argue for the importance of the notion of biological possibility by showing how it provides both a philosophically and biologically fruitful category as well as introducing a new practically grounded way for its assessment. More precisely, we argue that the new field of synthetic biology can provide tools to scientifically anchor reasoning about biological possibilities, and that in fact a significant portion of the field can be understood as investigating biological modalities.

Synthetic biology is a multidisciplinary field of biology that aims to take an engineer's viewpoint on biological systems and on the construction of biotechnological innovations. Its emergence has made the philosophical understanding of biological possibilities a more relevant problem. First, much of its inquiry is directed towards the domain of unactualized organisms (Elowitz & Lim, 2010). Second, synthetic biology itself provides tools to gain insight into biological possibility and to make modal inferences about biological systems. Synthetic biologists can explore the boundaries of biological constraints and design systems that overcome some of the limitations of naturally evolved organisms (Morange, 2009).

We distinguish two different strategies how this can be brought to bear on biological possibility: the *design* of functionally novel biological systems and the *redesign* of naturally occurring systems and their parts. We discuss two methods, rational design and directed evolution, as cases of design and redesign approaches, and assess how they are able to bypass some limitations of natural evolution. Applying these approaches allows synthetic biologists to explore designs that are not naturally evolutionarily accessible and draw inferences about relevant modal facts that go beyond the synthetic model systems themselves. Thus, we argue that synthetic biology has the potential to shed new light on various sources of unactualized possibilities in biology, like the historicity of generatively entrenched traits.

We begin the article in Sect. 2 by giving an introduction to the current state-of-the-art of biological modalities in the philosophy of biology. The main focus

is on the notion of biological possibility and its challenges. In Sect. 3 we describe synthetic biology, how it approaches biological systems, and discuss the role of modal claims in synthetic biology. Section 4 explores the specifically modal implications of synthetic biology practice, showcasing how different design methodologies allow for different kinds of modal inferences. We also critically assess these methods and highlight their limitations before concluding the article in Sect. 5.

2 Biological modalities

Traditionally, a large portion of biological research has approached its objects of study from a *descriptive* perspective. Due to the immense diversity of nature, much biology has focused on cataloguing, describing and systematizing biological species and the different forms that organisms take. This variety of organisms past and present is often conceptualized in a unified fashion under the metaphor of a single overarching *tree of life* (e.g., Darwin, 1859). The tree of life describes the ancestral relationships of organisms and thus their evolutionary history, each species forming their own sub-branch somewhere within the tree's branches.

Consequently, the tree of life metaphor describes the forms that life has taken in its actual evolutionary history. But descriptively and taxonomically oriented biological research is less adept to answer hypothetical "what if things were different" questions. These counterfactual questions, however, are indispensable when trying to assess such things as the contingency of certain traits, the plausibility of alternative evolutionary pathways, the causal specificity of genes or the necessary conditions of life (e.g., Dennett, 1995; Weber, 2017). The same is true in the case of scientific modeling more generally, as any philosophical account of modeling practices needs not only to accommodate the actual but also "grapple with the importance of consideration of the merely hypothetical" (Godfrey-Smith, 2014: 22). Questions like this urge researchers to better understand the nature of *biological modality*: what is possible, and what is not, in the domain of biological systems.

Considering how important the notion of possibility is, there is surprisingly little discussion that explicitly aims to tackle biological possibility. Several research areas in biology do deal with modal statements related to possibility, either directly, as in the case of evolutionary contingency,¹ or indirectly, as in the case of constraint and convergence. However, the concept of biological possibility itself has received relatively little attention in the philosophy of science. This might be partially based on the relative weakness of biological generalizations. It is generally understood that compared to physical and chemical laws, biological generalizations are significantly weaker: they are susceptible to exception, are domain-specific and might only hold *ceteris paribus* (Raerinne, 2013). In other words, biological generalizations do not exhibit enough regularity and universality to be considered law-like in a similar

¹ In classical modal logical accounts, a contingent statement is one that is possible, but not necessary (i.e., the statement's negation is also possible).

fashion as some generalizations in physics. The domain specificity and the lack of laws makes it more difficult to define clear boundaries of biological possibility. With this in mind, we will next describe how biological possibility is usually characterized and specify some features that an adequate account of biological possibility should have.

2.1 Framework for biological possibility

Generally, scientific modalities are conceptualized in a hierarchical manner, where certain modal claims are taken to be stronger and restrict the range of other claims (Dennett, 1995: 107). For example, however we are to understand the notion, what is biologically possible must also be physically possible. In addition, physical possibility also includes systems that might not be biologically possible. Within biology we can also discern different subtypes of biological possibility, for instance, genetic, developmental or evolutionary possibility. These types of possibilities do not form a similarly clear hierarchy between themselves. However, different biological possibilities are often interdependent of each other by providing several constraints to the biological system. For example, the developmental trajectory of an organism is not completely defined by its genetic makeup but is highly dependent on it.

Despite the interdependence of different types of biological possibility, the most developed account of biological possibility is defined in genetic terms, giving genetic possibility a more privileged role. This has several reasons. First, genes play a (sometimes unwarrantedly) fundamental role in biological thinking and the genetic level is seen as the preferred level of explanation (Rheinberger & Müller-Wille, 2017). Second, the same genetically based framework can be applied to all organisms, as the genetic code provides one of the most ubiquitous and regular features of organisms. Third, by defining biological possibility through DNA, one can also make its connection to chemical and physical constraints more evident, as many biologically functional features of DNA are dependent on the chemical and physical features of its structure and chemical constituents. And finally, the genetic framework provides a sense of conceptual clarity and tractability, as the number of differences between two genetic sequences can be relatively easily compared. In *Darwin's Dangerous Idea*, Daniel Dennett (1995: 118)² gives the following definition of biological possibility:

(BP) x is biologically possible if and only if x is an instantiation of an accessible genome or a feature of its phenotypic products.

Dennett's conception is based on the so-called *Library of Mendel* idea of the set of all DNA sequences. All known organisms form only a tiny subset of this astronomically large genetic space with an overwhelming majority of sequences corresponding to biologically impossible beings or to contingently unactualized possibilities. What is possible is determined by one's starting point in the genetic

² See also Huber (2017), who develops a formally rigorous and expanded presentation of the Dennettian framework to biological possibility by utilizing tools from modal logic.

space, rendering some outcomes more possible than others. The notion of accessibility is identified with moves that increase fitness or are at least neutral. In principle, however, the Library of Mendel should contain within itself the entire set of biologically feasible DNA-based organisms, the so-called abstract biological *design space* (Dennett, 1995: 125).

Dennett's account provides a convenient starting point for discussions on biological modality. Any notion of biological possibility that aims to capture central characteristics of modal claims in biology should fulfill a few features.³ First, there should be some definition of accessibility between two possibilities. This allows one to make claims about the possible trajectories of biological systems, about their robustness, and how these trajectories are constrained, for example, due to fitness concerns. Second, any notion of possibility should be relativized, depending on the starting point, and on the available methods how other biological possibilities can be accessed from that starting point. Third, modal claims should be able to vary in degree: some outcomes or structures are more possible than others (see Beatty, 2006). This does not necessarily entail that different possibilities can be assigned a definite probability, but only that different outcomes should be comparable in principle and the definition of biological possibility should define what it means for one outcome to be more possible than another.

However, even though Dennett's account satisfies in principle these three desiderata, it is rather abstract and far-removed from scientific practice to really give us tools to appraise specific possibility claims. Rather, Dennett's definition of biological possibility is descriptive and analytic in its outlook in the sense that it can account for most modal inferences only after the relevant biological details have been worked out. For example, we rarely have the knowledge to predict what kinds of moves are in fact evolutionarily accessible. What counts as a starting point in a genetic space and which kinds of moves can be deemed accessible changes in the course of evolution. Dennett (1995: 76), writing in the early 1990s, also anticipated the ability of biological engineering to reshape our understanding of these basic cornerstones of biological possibility. Dennett's account is best understood as a sketch of a framework that shows that biological modalities *can* be reasoned about without succumbing to Quinean worries about the epistemic opacity of possible entities and their properties (see Quine, 1953). However, to account for the wealth of modal reasoning in actual biological practice, it must be amended with a more fine-grained analysis that tells why huge parts of the biological design space remains unactualized.

2.2 Sources of unactualized possibilities

In biology, there are at least three possible ways how natural evolution fails to explore design space exhaustively. These three sources of unactualized possibilities are weak natural selection, lack of environmental variation and historicity

³ Wong (2020) provides a partially similar analysis on the desiderata of evolutionary contingency accounts.

(Wong, 2019).⁴ If natural selection is *weak*, then it is unable to explore the space of possibilities efficiently. Consequently, weak natural selection is more likely to fix on local maxima and less likely to find more globally optimal solutions. Second, the space of actualized biological systems might be limited due to the lack of environmental variation. As the evolutionary path of a population is dependent on the selective pressures of its environment, the set of actualized biological possibilities depends on the available environments which populations face during their evolution. And third, historicity of organisms constrains possible future evolutionary paths. The space of possible evolutionary paths that a species can take tends to narrow during its evolutionary history. When evolutionary traits become fixed, they constrain the ways in which the organism might evolve further. Evolution, as Francois Jacob (1977) notes, is a tinkerer, working on the material it has access to, and modifying existing structures rather than producing *de novo* solutions. As a general trend, organisms become more integrated and complex through evolution, and traits tend to persist even after becoming useless and atrophied.

Even though some traits seem to be fixed and ubiquitous in a range of organisms, their emergence might have itself been a contingent event. This phenomenon is called *generative entrenchment*. Generative entrenchment is a concept introduced by Schank and Wimsatt (1986) and describes a phenomenon where traits become fixed in development and evolution due to the irreversible nature of their emergence. However, this does not necessarily entail that the trait itself is optimal or the only possible way of realizing the relevant functionality. Consequently, one cannot presume necessity just because some trait is commonly found in multiple taxa. A classic example is the ubiquity of the genetic code. Although not dictated by its underlying biochemistry, the particular mapping from DNA sequence to amino acids is virtually universal on Earth, making some deem it a “frozen accident” in the history of life (Schank & Wimsatt, 1986).

Generative entrenchment promotes stability by introducing evolutionary inertia: as changes in generatively entrenched traits are more likely to lead to system failure or at least negative fitness outcomes, these traits become conserved in evolution. Or, as Desjardins (2011: 241) describes it, generative entrenchment “has a tendency to stabilize certain regions of the evolutionary state space.” Backtracking such traits once they become fixed is impossible for natural selection. But other methods beside natural evolution can be used to study the extent of entrenchment of the traits, their optimality conditions and even produce alternatives to naturally fixed traits. We will next turn to describe synthetic biology and how it can contribute to the understanding of biological possibility.

⁴ Wong (2019) explicitly discusses the sources of evolutionary idiosyncrasies, that is, cases where natural evolution fails to generate the same form (such as the platypus) more than once. We consider this analysis also applicable to several unactualized possibilities. One additional obvious source of “unactualized possibilities” is sub-optimality: cases when natural evolution does not produce some outcome because it is selected against.

3 Synthetic biology and biological possibility

The website *syntheticbiology.org* defined synthetic biology as “the design and construction of new biological parts, devices, and systems and the redesign of existing, natural biological systems for useful purposes” (in Calvert, 2010: 96). This definition points to some features that are important when analyzing how synthetic biology relates and contributes to the understanding of biological possibility. First is the distinction between the *design* of novel organisms and the *redesign* of existing living systems. Although these two approaches are not completely independent avenues of research, they have important differences and frame the relationship between synthetic biology and natural organisms from distinct perspectives. We will discuss these differences, as well as how design and redesign approaches are used to understand biological possibility, more thoroughly in the next section.

Second, the definition provides a distinction between natural biological systems and those constructed and studied by synthetic biologists. This “divergence of nature” principle, as Schmidt (2015: 6) calls it, contrasts synthetic biology with the natural world, and nature works as a “central anchor and negative foil for this definition”. As Pade et al. (2015) argue, these novel non-natural functionalities are one of the main characteristics of synthetic biological entities. They cite the *New and Emerging Science and Technology High-Level Expert Group’s* definition of synthetic biology, where they define synthetic biology as bringing about “functions that do not exist in nature” (NEST, 2005 in Pade et al., 2015: 76).

Synthetic biology expands the domain of living things by constructing novel synthetic organisms that diverge from the functionalities of naturally evolved organisms.⁵ We distinguish four different motivations for synthetic biology to explore biological possibilities: improvement of nature, novel functions with human benefits, gaining understanding about nature and biosafety. First, synthetic biologists might want to improve designs of natural evolution. This need for improvement is motivated by the claimed sub-optimality of the natural biological system. Proponents of synthetic biology have on several occasions claimed that one of the benefits of synthetic biology is its ability to overcome the limits of natural selection and provide better designs than those available in natural systems (e.g., Endy, 2005). However, it should also be noted that synthetic biology does not necessarily aim to produce evolutionarily optimal solutions. This is especially obvious in cases that are motivated by the second type of reason to explore biological possibilities: the design of new-to-nature functional systems. These synthetic systems exhibit novel functionalities defined by the designer that are beneficial for human purposes. Synthetic organisms might be constructed to give economic or health benefits for humans, providing a variety of agricultural, industrial and biomedical applications, such as biochemical sensors that detect pathogenic states and activate treatment-promoting metabolic

⁵ However, currently real novel synthetic organisms have only been achieved in microbiology through the manipulation of bacteria (Gibson et al., 2010) and yeast (Richardson et al., 2017). Beyond some extreme forms of protocell engineering (Solé et al., 2007), practically all synthetic biology operates with a select number of already existing laboratory organisms.

pathways (Weber & Fussenegger, 2012). In the design of new-to-nature functional systems, the actual ecological and selective feasibility of the target system is typically of little interest or is even totally disregarded, save for growth and performance in a carefully controlled lab environment. In these cases, synthetic organisms might have functionalities that produce no benefit for the organism itself, and, furthermore, decrease the fitness of synthetic organisms compared to their natural counterparts. For example, the addition of a synthetic DNA device to an organism can decrease its growth rate through metabolic burden by requiring additional RNA molecules and proteins, or by interfering with native cellular processes (Renda et al., 2014). This imposes a problem to synthetic biologists, as deleterious mutations that suppress the designed functionalities are selected for, and synthetic systems lose their reliability after a certain number of host generations.

These two ways of exploring biological possibilities, by improving sub-optimal nature or producing novel functionalities, are usually taken as the most prominent motivations of synthetic biology. The construction of new or improved organisms for human benefits offers the most immediate promises of a new field. It should be noted that these motivations are not methodologically and conceptually completely distinct: although synthetic biologists might talk about “improving nature,” what they actually mean by it is improving it for our purposes by producing systems with new-to-nature functions. Furthermore, one could reasonably describe the optimization of a natural metabolic pathway through heterologous pathway design both as a process of improvement or as novel design (e.g., Yadav et al., 2012).

This focus on the applications, however, downplays other important contributions of synthetic biology and could make synthetic biology seem just an extension of biotechnology with more powerful tools. Besides practical applications, synthetic biology is also often used for more traditional “basic science” purposes, where synthetic systems are designed and built to gain understanding on natural systems (Knuutila & Loettgers, 2013). This is the third motivation for investigating biological modalities: to provide insight into biological systems and phenomena by manipulating and building biological systems. Often this is done by reducing the complexity of natural design, for example, by increasing modularity, reducing natural systems to well-defined parts or by removing genes.⁶ This avenue of synthetic biology has provided insight, e.g., on the role of noise in biological systems (Knuutila & Loettgers, 2014), biological design principles (Gramelsberger, 2013), and gene essentiality (Hutchison et al., 2016). And finally, synthetic biology might make their systems differ from natural organisms for biosafety reasons. Accidental release of synthetic organisms outside the laboratory or their intended domain of application is an acknowledged concern. However, it is suggested that synthetic organisms could be designed to be divergent enough from natural organisms to prevent this danger. For example, synthetic biologists have constructed mutant lineages that require non-naturally occurring nutrients (such as non-canonical amino-acids or synthetic

⁶ Besides basic science purposes, reduction of complexity is often also motivated by the desire to make organisms easier to engineer (Porcar & Peretó, 2016). This includes the attempts to create a toolkit of well-characterized parts for the rational design, which we will discuss in Sect. 4.

coenzymes) to survive, thus preventing the synthetic organism from being viable in nature in the case of accidental release (Torres et al., 2016).

We argue that a significant part of these synthetic biology endeavors can be understood as the investigation of biological modalities. There is a very straightforward interpretation that links synthetic biology and the investigation of biological possibility. This is based on the fact that whenever synthetic biologists construct new biological systems that diverge from naturally evolved organisms and their parts, in all likeness, they have actualized something new and biologically unique (see Kendig, 2016: 743). In synthetic biology, claims about biological possibility are not only counterfactual hypotheses. Rather, they provide a methodological framework and rationale for the design and redesign of synthetic systems. For example, it has been argued that the use of how-possibly models in synthetic biology differs from its standard explanatory-evidential use in mechanistic contexts, because synthetic biologists also aim to materially realize these models and use them to gain modal understanding (Koskinen, 2017).

It is an elementary feature of modal reasoning that every actual thing must also be possible (for if it were impossible, it simply couldn't be actual to begin with). This can easily be turned into a methodology for modal investigation. If we are unsure about whether a thing is possible or not, one epistemically powerful way to settle this issue is to try and actualize that very thing. If we succeed, we have managed to give a proof of a possibility. However, this simple methodological point is just a starting point in how synthetic biology can help us with our modal understanding. For technically, the said methodology is logically valid only for the particular token systems that have been constructed. It cannot guarantee any modal truths whose scope goes beyond those single laboratory products. But biologists are most of the time interested in types, not tokens; generalizable scientific knowledge rather than anecdotal facts. This is where the skill of researchers to make careful and cleverly constructed *modal inferences* comes into play. We argue that there is an important connection between the ability to make modal inferences (and the quality of the said inferences) and the use of certain design setups that we will turn to characterize next.

4 Modal inferences in synthetic biology

One of the main contributions of synthetic biology to the understanding of biological possibility, is due to its ability to overcome some constraints that restrict the work of natural evolution. As we discussed in Sect. 2, biological possibilities are often framed as forming some type of restricted design space, where natural organisms represent only a small subset of all possible organisms. Weakness of natural selection, lack of environmental variation and historicity all limit natural selection's capability to explore design space.

Whereas natural evolution is restricted by the topology of the organism's fitness landscape and tends to follow more immediately adaptive paths, synthetic biology provides a new way to explore design space, overcoming some of these limitations.

This does mean that synthetic biology is an unconstrained and superior method to access biological possibilities. Although synthetic biology is sometimes presented as a way to “improve nature,” it is in many ways more limited and constrained than natural evolution. Rather, synthetic biology is restricted in *different* ways than natural evolution, and that allows it to access unexplored areas of design space and to gain modal insight into biological possibilities. For example, synthetic biologists can try to explore the limits of embedded traits by synthesizing alternatives to various regulatory motifs (Elowitz & Leibler, 2000) or even the genetic code itself (Chin, 2017). Or, they can harness microorganisms for industrial purposes, building devices that utilize biological solutions, but take these completely out of any kind of natural evolutionary context. The success of these attempts to explore design space is highly dependent on the chosen design methods and their given heuristics.

4.1 Design and redesign as strategies for exploration

Synthetic biology is commonly defined through a two-branched definition that recognizes two approaches to constructing synthetic organisms: design of new systems and parts, and redesign of already existing ones (see Calvert, 2010). As we mentioned earlier, these two approaches are not completely independent and distinct: design and redesign activities often complement each other. Natural organisms can be incrementally redesigned to exhibit completely novel functionalities (Haseltine & Arnold, 2007). It has also been argued that very few constructs of synthetic biology can be described as thoroughly new. In truth, they are often for the most part modelled after natural organisms and even the most famous cases of organisms with wholly synthetic genomes should be described as cases of “genome plagiarism” (Porcar & Peretó, 2016: 451).

However, despite the occasional ambiguity, we do think that the distinction between design and redesign allows us to discern some interesting points on how these two approaches compare to natural evolution as a method of exploring design space. For the purposes of our analysis, we define design as the activity of constructing systems and devices with novel functions. One of the most prominent examples of this strategy is rational design, an engineering-inspired method which proceeds through bottom-up construction from well-defined standardized parts (Cambray et al., 2011). As discussed earlier, evolutionary development of organisms tends to lead in time to increased complexity (though there are exceptions to this general trend). This complexity manifests itself both in the higher number of relevant traits, parts and interactions, but also in the increased integration of system design, decreased modularity and stronger coupling between organism and its environment. Rational design allows synthetic biologists to reduce biological complexity and simplify their systems through the use of modular parts, and by designing systems to be more engineerable (Ijäs, 2019). One of the main goals of rational design synthetic biology is to separate natural systems to simpler well-defined parts that can be manipulated and recombined with predictable outcomes.

This design of new-to-nature functions is contrasted with the strategy of redesigning already existing organisms. We consider redesign strategy including two

conceptually distinct ways of approaching natural biological systems: as a starting point for further modifications, or as a model for alternative realization. We call these *functional* and *structural redesign*, respectively. Functional redesign is seen in cases where synthetic biologists use, for example, evolutionary methods such as directed evolution to optimize and reconfigure natural organisms (Romero & Arnold, 2009). As a design method, directed evolution is a form of artificial selection; deliberate mutations are first induced in the target, and desired phenotypes are then selected by the designer. This cycle between mutant generation and selection phases is iterated until a desired endpoint is reached (Marguet et al., 2007). In these respects, redesign through directed evolution resembles natural selection. However, as the direction of the process is decided by the human designer and not by the natural selective pressures of the organism, this allows it to reach areas that are inaccessible in the natural environment. With artificial selection methods such as directed evolution, the “power of evolution is thus harnessed as a strategy to synthesize biological possibilities” (Simons, 2020: 17). In turn, an example of the structural redesign comes from the study of alternative genetic systems. Here the natural function of DNA is taken as a goal, but the structural realization to attain that function is changed. Partial success has thus far been met with foreign building blocks like new nucleobases in place of A, C, G and T, as well as codes that utilize novel sequence-amino acid mappings compared to the natural code (Chin, 2017).⁷

Both rational design and directed evolution have their characteristic limitations. In the case of rational design, as new synthetic systems are constructed bottom-up almost from scratch, it requires synthetic biologists to have access to a toolkit of relevant well-defined biological parts and devices. Synthetic biologists also require detailed mechanistic understanding of the systems that they are constructing (see Ijäs, 2019). In turn, for directed evolution to access a certain outcome, there must be a pathway of incremental detectable beneficial mutations from start to endpoint (Dougherty & Arnold, 2009). Directed evolution is, therefore, affected by the topography of the fitness landscape and by the designer’s ability to screen for desired independent properties.

We can compare these two design and redesign methods, rational design and directed evolution, on how they are capable of overcoming the effects of previously discussed sources of unactualized possibilities: weak selection, lack of environmental variation and historicity. The effects of weak selection and lack of environmental variation do not really apply to rational design, as its design method is qualitatively different from evolution. But its exploration of design space has its limits. Compared to redesign methods such as directed evolution, rational design should at least theoretically allow the designer to take qualitative leaps in the design space and construct something completely novel (Haseltine & Arnold, 2007). However, the more novel or farther away from current organisms the synthetic system is, the harder it is for the synthetic biologist to have the required understanding to construct this new

⁷ The distinction between structural and functional redesign is not categorical. For example, artificial genetic codes are often built not only to reprogram the extant code mapping, but also to expand it beyond the 20 canonical amino acids found in nature.

biological possibility. One could argue that rational design can make these “huge leaps through design space” (Dennett, 1995: 76–77) creating organisms that would never have evolved by ‘ordinary’ means, but only when the endpoint is already known. This is further hampered by the effect of context-sensitivity to rational design: biological components and devices are susceptible to losing their functionality upon reaggregation or moving to another environment (Ijäs, 2019). Consequently, many parts of the rational design toolkit have limited functional ranges, a factor that limits the “free exploration” of design space.

In turn, the power of directed evolution can be quite easily compared to natural evolution. Like natural evolution and in contrast to rational design, directed evolution explores biological possibilities more continuously, without obvious qualitative leaps. Their exploratory capability is therefore defined and limited by the selection process and the available variation (Dougherty & Arnold, 2009). Directed evolution is somewhat more capable of overcoming the limitations of weak natural selection. The selection applied in the directed evolution design cycle is very strong, as usually all non-desired phenotypes are selected or screened off. However, the availability of mutational variation is a bigger obstacle. Previous uses of directed evolution, like traditional artificial selection, can be described as proceeding through “strong selection, weak mutation,” although new library generation methods have increased the available variation in each design cycle (Currin et al., 2015). In addition, directed evolution is also affected by the topography of the fitness landscape and is, like natural selection, susceptible to stop at local maxima (Romero & Arnold, 2009).

Success of directed evolution is, therefore, to a large extent determined by the fruitfulness of the starting point from where the design cycle is initiated. Consequently, it is, like natural evolution, constrained by the third source of unactualized possibilities, historicity. It is difficult to backtrack or decouple entrenched traits via artificial selection alone. Other design and redesign methods are more suitable for these tasks, as we will shortly discuss. Directed evolution could probably be used to study the contingency of evolutionary events by taking a precursor system as a starting point and then guiding it to evolve into an alternative pathway. However, we would suggest that more insight into these types of questions concerning the evolutionary contingency of certain traits could be gained instead by using experimental evolution, as suggested by Desjardins (2011).

Synthetic biology can also provide us with new insight on a specific case of historicity, namely generative entrenchment, which we discussed earlier in Sect. 2. One reason why traits become fixed is a phenomenon that Schank and Wimsatt (1986) call *accretion*, which is the accumulation of new traits in the later stages of evolution. These traits are interconnected and possibly even dependent on the generatively entrenched trait. This increases the integration and non-modularity of the system, at least for the entrenched traits that cannot be intervened without drastic changes in the system. Subsequently, changes in the entrenched trait would require one first to wind down several other traits that have accumulated later. Otherwise, interventions in the generatively entrenched trait might, due to the interconnectedness and centrality of the trait, cause changes that propagate through the system, leading to unpredictable qualitative changes or probably total system failure. However, as we mentioned earlier while discussing the design approaches, one of the central tenets

of synthetic biology is to increase modularity by simplifying biological systems through decoupling and hierarchical abstraction (Endy, 2005). Synthetic biologists might either aim to create a new system *de novo* bottom-up, where entrenched traits are changed, or to reduce a natural system by removing accumulated parts to decrease integration and complexity. This might allow synthetic biologists to understand the real contributions that generatively entrenched traits make, and whether they can be considered adaptively optimal.

Due to generative entrenchment, the theoretical existence of functionally equivalent (or better) solutions does not mean that these can be accessed by selection as there would be a significant loss of function in between the evolutionary steps (Wagner, 2005). However, we could still be interested to know whether these systems correspond to something biologically possible. Directed evolution is unlikely to be of much use here. However, in contexts like this, structural redesign employing rational design methods is likely to carry the greatest potential. By keeping the functional goal fixed, this approach circumvents some of the problems of purely free-form exploration of the unknown ranges of the design space while at the same time not being as bound by selective pressures of immediate fitness benefit (Koskinen, 2019). Examples come from labs working with alternative realizations of biological functions, like experiments with novel genetic codes or “mirror life” utilizing biomolecules of non-natural chirality. Some of these systems could also find application in biosafety, as their material orthogonality would ensure a biological firewall between them and naturally occurring systems (Torres et al., 2016).

4.2 Discussion

Application of synthetic biology design allows one to access naturally inaccessible areas and get a better sense of the constraints affecting them, namely which areas are accessible via genetic engineering and which are strictly constrained or bounded by trade-offs. Ollé-Vila et al. (2016: 487) invoke Francis Bacon’s metaphor of interrogation of nature to describe this exploration of empty areas of design space with synthetic biology techniques. The relationship between design and redesign can be partly understood as a difference between stances to the kinds of modal reasoning that the target systems are taken to exhibit. For example, harnessing the cellular machinery of microorganisms for human purposes investigates the industrial potential of biology. While the resulting systems would still technically occupy the same abstract design space as naturally occurring organisms, they could end up being selectively nonsensical. That is, no interpretation of evolutionary accessibility would likely make sense for them. In contrast, possibilities studied through redesign methods are often partly framed through evolutionary contingency: is the prevalence of a particular trait a functional necessity or a historical accident?

Making modal inferences is not in any way an easy endeavor, not least in the context of messy and complex biological systems. From a scientific standpoint, actual experiments are often riddled with various uncertainties and underdetermined options for future theory-building. However, conceptually speaking, modal notions, being related by definitional stipulations, provide richly interconnected inferential

avenues that are in theory simple to navigate. While a possibility can be inferred from an actuality, different carefully selected design choices allow for further, more fine-grained deductions. For example, two mutual possibilities for the realization of a trait suggest *contingency*. The inference is suggestive as it does not yet tell us about counterfactual histories directly: whether life *could have* evolved in another way. But it is a start, and can help us better evaluate the plausibility of such hypotheses. In turn, structural *necessities* arise when there is essentially only one viable way to build the said function.⁸ *Impossibility* claims have been traditionally problematic cases in the philosophy of science, since there is rarely a direct route to prove that something is not possible. Any number of token failures to realize a system of particular design cannot logically guarantee that the said system is strictly speaking impossible. However, embedded in a larger context of biological knowledge, e.g., as informed by quantification of viable design in systems biology (Green, 2015), conscious design choices in synthetic biology can make it easier to reveal tacit constraints that help us understand why some system features might remain, if not impossible, at least *highly unlikely*.

The resulting synthetic systems are thus best understood not just as singular artifacts with potentially useful practical applications, but as highly theoretically motivated model systems that figure as vehicles of reasoning (Knuuttila & Loettgers, 2013). However, how these synthetic biological models are used to reason about actual and possible biological systems, should be done with a certain caution in mind. What kind of insight can we gain about biological organisms and their possibilities by constructing and modifying novel synthetic systems? This is one of the ambiguities of using synthetic biology to understand life (see Simons, 2020). As Evelyn Fox Keller (2002: 288) notes, synthetic organisms “are still simulations, albeit in a different medium.” Synthetic biologists often invoke Richard Feynman’s famous quote “What I cannot create, I do not understand” as a motivation for their endeavor (e.g., Marguet et al., 2007). However, if synthetic biology provides models for nature, one should consider whether the model systems really provide understanding on nature, or only on the synthetic systems themselves. With this in mind, we have in this paper tried to outline some ways in which synthetic biology can be used to understand biological possibility and what considerations should be taken into account when applying these methods to make modal inferences.

5 Conclusions

In this paper we have offered an analysis of the interplay between the emerging field of synthetic biology and the notion of biological possibility. We have argued for two main points about the role of modal claims in synthetic biology. First, we have suggested that a significant part of synthetic biology can be understood as inquiry about biological modalities: what is possible, and what is not, in the domain of biological systems. Second, we have specified several ways how synthetic biology can provide new tools and insight concerning the study of biological possibilities. Previous

⁸ Dennett calls these kinds of situations *forced moves* in design space (Dennett, 1995: 129).

accounts of biological possibility have provided a useful conceptual basis for the basic semantics for biological modal statements (Dennett, 1995; Huber, 2017). However, synthetic biology's exploration of novel biological systems can concretize the more speculative aspects of biological possibility, and also expand our understanding of its scope. We have argued that there is an important connection between the ability to make modal inferences and the use of particular design setups we have identified and characterized. This helps us to understand the kinds of stances synthetic biologists take towards biological possibility and see how different kinds of modal inferences are justified. While novel synthetic systems often realize previously inaccessible possibilities, care must be taken when determining the scope of modal facts that these can be taken to instantiate. This is where the skill of the researchers to make careful and cleverly constructed modal inferences comes into play.

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Declarations

Conflicts of interest Both authors declare that they have no conflicts of interest.

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