Genetic frontiers for conservation

An assessment of synthetic biology and biodiversity conservation

Edited by: Kent H. Redford, Thomas M. Brooks, Nicholas B.W. Macfarlane, Jonathan S. Adams

INTERNATIONAL UNION FOR CONSERVATION OF NATURE

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About the IUCN Task Force on Synthetic Biology and Biodiversity Conservation

The IUCN Task Force on Synthetic Biology and Biodiversity Conservation and its accompanying Technical Subgroup were put together to accomplish the tasks laid out in Resolution WCC-2016-Res-086 from the 2016 World Conservation Congress. This Resolution (in part) called on the Director General and Commissions to undertake an assessment to:

examine the organisms, components and products resulting from synthetic biology techniques and the impacts of their production and use, which may be beneficial or detrimental to the conservation and sustainable use of biological diversity and associated social, economic, cultural and ethical considerations…

In addition, it called upon the Director General and Commissions with urgency to:

assess the implications of Gene Drives and related techniques and their potential impacts on the conservation and sustainable use of biological diversity as well as equitable sharing of benefits arising from genetic resources…

This assessment is the result of the work of the Technical Subgroup managed by the Task Force. Both the Task Force and the Technical Subgroup were established in January 2018.

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Cover photo: Copyright Shutterstock/Enrique Aguirre. Working with the tools of synthetic biology will present conservation with a number of challenges and opportunities that will ripple across the natural world, reaching even places like the Andes Mountains with their iconic guanacos.

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Foreword

The explosion of knowledge that research on DNA has brought has been extraordinary. The recent, rapid development of gene sequencing and editing technologies has led to the creation of a new generation of tools. The technologies that allow humans to alter the genes of organisms to make them do things that humans want and that those organisms would not normally do -- for example, creating yeast that can make plastic or human medicine -- is called synthetic biology. There is an active international discussion on how best to define the field.

Scientists now have tools available that in principle may allow them to make changes to the genetic makeup of nearly every species, including, but also extending well beyond, single gene manipulation. DNA can be copied into digital form, rearranged, turned back into organic form, then inserted back into living cells in an attempt to strengthen or create desirable characteristics or eliminate problematic ones. These new and rapidly evolving technologies create exciting opportunities in many fields, including new kinds of conservation, but they also raise serious questions and complex challenges.

It was both deep concern and qualified excitement that led IUCN to commission a broad assessment of the current state of science and policy around synthetic biology techniques as they relate to biodiversity. The goal of this assessment is therefore to provide a clear understanding, based on the best available evidence, of the issues regarding synthetic biology that are relevant to and may have an impact – positive or negative – on the conservation and sustainable use of biological diversity. Produced by a global team of practitioners and researchers, this assessment responds in part to an IUCN Resolution adopted at the IUCN World Conservation Congress in 2016: "Development of IUCN policy on biodiversity conservation and synthetic biology" (WCC-2016-Res-086).

Application of synthetic biology to conservation is in its earliest stage. That makes the requirement that this assessment use an evidence-based approach more challenging but even more vital. While policy debates necessarily engage values and preferences, claims in support of, or in opposition to, synthetic biology that draw primarily from these need to be distinguished from those grounded in evidence. This assessment thus aims to shed light on the state of the field, with the potential benefits and harms discernible to date. It cannot be, and does not aim to be, a comprehensive risk assessment. Rather, the goal of this assessment is to inform future deliberations and increase the understanding of the different ways that evidence regarding the potential impact of synthetic biology on conservation is generated, used, and interpreted.

This assessment is the beginning of a process that will lead to the development of an IUCN policy to guide the Union's Director General, Commissions, and Members. The draft policy will be discussed in many fora before it is brought to vote at the World Conservation Congress in 2020. Far greater public attention to the topic of synthetic biology and biodiversity conservation is essential, given the potential impact of scientific discoveries and policy decisions that may be just over the horizon, and also given the need for broad partnerships to address the challenges that the conservation and synthetic biology communities will inevitably face.

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Statement of principles of the IUCN Task Force on synthetic biology and biodiversity conservation

Recognising the complexity and large positive and negative potential impacts of the subject, both on and beyond the global conservation community, this assessment will draw on the values and proven processes of IUCN to provide a shared and trusted resource for subsequent deliberations.

In preparing the assessment on behalf of the IUCN membership, the Technical Subgroup has striven to adhere to the principles of:
Objectivity – assessing evidence and working to minimise and balance subjective bias;
Inclusivity – recognising and being considerate of the full diversity of views and interests;
Robustness – ensuring that all conclusions drawn are based on clear reasoning;
Humanity – interacting with all interested parties in a respectful and honest manner;
Transparency – ensuring that the process applied and all final outputs arising from it will be open access;
Consultation – giving meaningful opportunities for all interested parties to engage with the process, and responding to all formal submissions.

The work is all conducted under the umbrella of the IUCN Commission Code of Conduct and the IUCN Secretariat Code of Conduct.

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Glossary

See Box 1.1 for standard introductory genetics terms.

Allele: a form of a gene at a particular position (locus) on a chromosome.

Autosome: chromosomes which are not sex chromosomes (such as X and Y in mammals).

Bioaugmentation: the addition of archaea or bacterial cultures required to speed up the rate of degradation of a contaminant.

Biodiversity: biological diversity, "the variability among living organisms from all sources including, inter alia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part; this includes diversity within species, between species and of ecosystems" (CBD 1992).

Bottleneck (population): an ecological event that drastically reduces a population producing evolutionary impacts.

CITES: Convention on International Trade in Endangered Species of Wild Fauna and Flora. It is an international agreement between governments aimed at ensuring that international trade in specimens of wild animals and plants does not threaten their survival. It entered into force in 1975, and currently has a membership of 183 Parties.

CRISPR-Cas9 technology: biochemical method using clustered regularly interspaced short palindromic repeats (CRISPR) guide RNA in conjunction with Cas9 (CRISPR-associated 9) nuclease to efficiently cut and edit DNA.

De-extinction (or species revival): the development of functional proxies for species which have previously become extinct.

Digital sequence information on genetic resources: contested term referring to certain types of genetic information derived from DNA sequencing.

DNA sequencing: detecting the sequence of the four bases (adenine, thymine, guanine, cytosine) as the code of genetic information.

DNA synthesis: process of creating natural or artificial DNA molecules.

Functional genomic screening: a key discovery enabling the identification of gene and protein function.

Gene drive: A phenomenon of biased inheritance in which the ability of a genetic element to pass from a parent to its offspring through sexual reproduction is enhanced, leading to the preferential increase of a specific genotype that may determine a specific phenotype from one generation to the next, and potentially throughout a population. A gene drive element is a heritable element that can induce gene drive, such that the gene drive element is preferentially inherited. Gene drive elements may be referred to as gene drive systems or simply "gene drives."

Gene flow: exchange of genetic material between populations, either through individuals, or mediated through pollen, spores, seeds or other gametes.

Genetic drift: random change of genetic variation from one generation to another.

Genetically modified organism (GMO): also known as "living modified organism" (LMO), an organism whose characteristics have been changed by genetic engineering (contrasting classical selection experiments or naturally by mating and/or recombination).

Genetic rescue: deliberate introduction of individuals or gametes as vehicles for the infusion of novel alleles (hence to increase gene flow, genetic diversity and fitness).

Genome editing: making targeted changes to the genome of an organism, predominantly by using site-specific endonucleases such as CRISPR-Cas9.

Genotype: the genetic constitution of an individual organism.

Inbreeding depression: whereby the expression of deleterious recessive traits is more likely due to lower gene pool diversity, resulting in reduced fecundity and/or survival.

Invasive Alien Species: taxa that are introduced accidentally or deliberately into a natural environment where they are not normally found, with serious negative consequences for their new environment.

Mendelian inheritance: form of inheritance proposed by Gregor Mendel with the following laws: law of segregation, law of independent assortment, law of dominance. Characteristics are inherited from parents to offspring individuals following those laws in predicted ratios.

Pathogen: a biological agent that causes disease or illness to its host.

Phenotype: the ensemble of observable characteristics displayed by an organism.

Risk: The likelihood and severity of a potential adverse effect. For example, if the likelihood of an adverse effect occurring is high, but the severity of the adverse effect is very low, the overall risk will be low. If, however, the severity of the adverse effect is extremely high, even a low probability of it occurring may still be considered a large risk. That is, even if there is only a 1% chance that an approaching asteroid will destroy the earth, this will likely be considered a high risk that needs to be addressed.

Risk assessment: the structured process for analysing risk.

Recombination: In the process of transferring genetic information from parents to offspring, new combinations of traits can occur, caused by recombination of chromosomes during meiosis.

Release of insects carrying a dominant lethal (RIDL): release into the wild of insects carrying a dominant lethal gene or genetic system.

Selection: Some individuals in a population have higher reproductive success, as they possess characteristics which make them more adapted to their environment.

Squalene: a natural 30-carbon organic compound originally obtained for commercial purposes primarily from shark liver oil (hence its name, as *Squalus* is a genus of sharks).

SRY mice: Sry is a sex-determining gene that regulates testis differentiation; in SRY mice this gene is placed on an autosome and offspring are only male.

Sterile insect technique (SIT): a technique in which sterile individuals of a species are generated in the lab (e.g. through radiation) and then released into the wild.

Sterile male: Sterile males are released into nature such that, when mating with wild females, there are no offspring. Males are sterilised either through radiation or by genetic manipulation.

Symbiosis: any type of a close and longer-term biological interaction between two different biological organisms, be it mutualistic (benefits for both), commensalistic (benefits for one while no harm to the other) or parasitic (benefits for one while causing harm to the other). The organisms, each termed a symbiont, may be of the same or a different species.

Transgene: a gene or genetic material that has been transferred naturally, or by any of a number of genetic engineering techniques from one organism to the other. The introduction of a transgene (called “transgenesis”) has the potential to change the phenotype of an organism.

Vector: any agent that carries and transmits an infectious pathogen into another living organism.

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Tables

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1.
What does synthetic biology and gene drive have to do with biodiversity conservation?

Todd Kuiken, Edward Perello, Kevin Esvelt, Luke Alphey

Image by: SKHerb / Shutterstock.com

1.1 Introduction

The loss of Earth's biodiversity is accelerating at an unprecedented rate and proceeding at all levels: ecosystems, species and genes. No corner of the Earth, no matter how remote, is today free from human influence, whether in the form of the altered atmosphere, expanding cities, ubiquitous pollution and invasive species, conversion of wildlands and loss of once fertile farmland, or expanding exploitation and trade of wild species. Governments have set ambitious targets for addressing biodiversity loss worldwide, such as the Aichi Targets of the Convention on Biological Diversity's Strategic Plan 2011–2020, and the Sustainable Development Goals (SDGs) agreed by the United Nations in 2015 (UN, 2015). To date, however, both the targets and the institutional arrangements that support them are singularly failing (Tittensor et al., 2014).

In recent years synthetic biology has emerged as a suite of techniques and technologies that enable humans to read, interpret, modify, design and manufacture DNA in order to rapidly influence the forms and functions of cells and organisms, with the potential to reach whole species and ecosystems. As synthetic biology continues to evolve, new tools emerge, novel applications are proposed, and basic research is applied; much remains to be learned about which genes influence which traits and how they may interact with each other and with environmental factors, including via epigenetic phenomena (for a description of epigenomics, see Box 1.3). Much of synthetic biology innovation, especially in enabling technologies (Figure 1.1) is considered to be exponential, and it is considered a domain of the Fourth Industrial Revolution, blurring the lines between the physical, digital and biological spheres. The Industrial Revolution refers to the fourth major industrial revolution and is characterised by its "velocity, scope, and systems impact" and the combination of technologies from the physical, digital and biological realms (Schwab, 2016). The emerging capabilities, applied to the conservation of biodiversity, have great potential to reshape the conservation field in unforeseeable ways, both positive and negative and along unknown timelines.

This assessment is one part of IUCN's effort to provide recommendations and guidance regarding the potential positive and negative impacts of synthetic biology on biodiversity conservation. Past efforts and resolutions of IUCN have examined the impacts and potential uses of genetically modified organisms in relation to biodiversity (IUCN World Conservation Congress, 2000, 2004; Balakrishna, Dharmaji & Warner, 2003; Congress, 2004; Young, 2004). Taken together these will serve as an input to the development of policy recommendations to be debated and voted on by the IUCN membership at the 2020 World Conservation Congress in Marseilles.

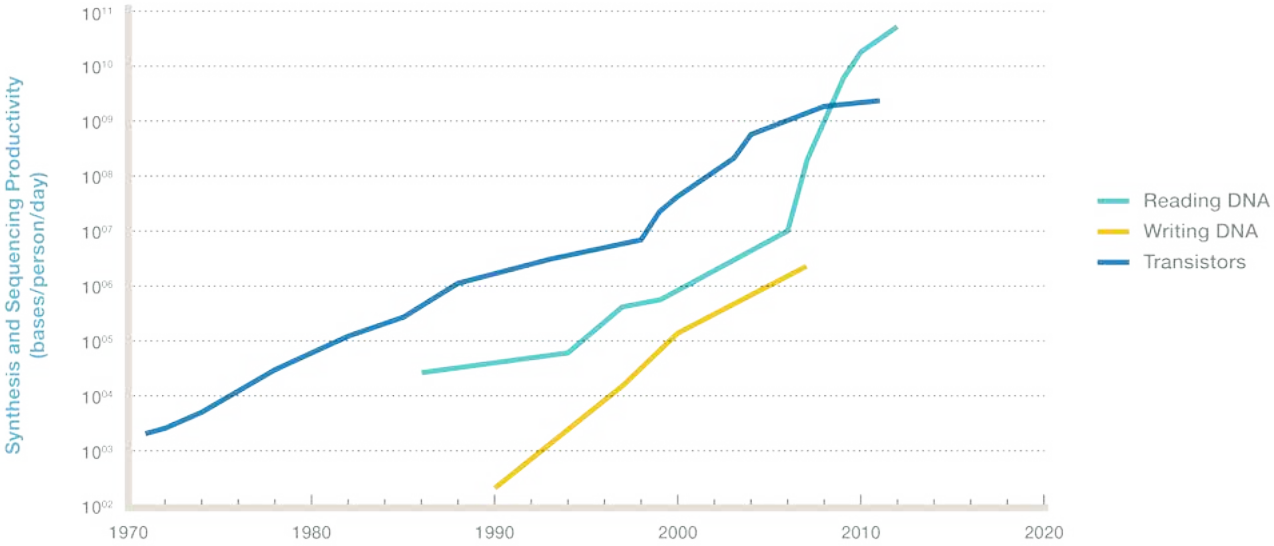


Figure 1.1 The productivity of DNA synthesis and sequencing, measured as bases per person per day, using commercially available instruments, and compared to Moore's Law, which is a proxy for IT productivity. Productivity in sequencing DNA has increased much faster than Moore's Law in recent years. Productivity in synthesising DNA must certainly have increased substantially for privately developed and assembled synthesisers, but no new synthesis instruments, and no relevant performance figures, have been released since 2008. Adapted from Bioeconomy Capital, 2018.

1.2 Interaction of the synthetic biology and biodiversity conservation communities

The emergence of synthetic biology has led to tension within the global conservation community and a growing understanding of the utility of deeper and more meaningful interaction between contemporary conservation and synthetic biology communities (Piaggio et al., 2017). The governments of many developing countries, indigenous leaders and local communities have also voiced concerns over how synthetic biology may affect their cultures, rights and livelihoods. Both the hopes and fears surrounding the application of synthetic biology to conservation stem from the same troubling observation: the loss of biodiversity continues despite the growing sophistication of conservation activity and conservation science; and the understanding among governments at all levels as well as civil society that human well-being depends on a thriving natural world.

For some in the conservation community there is sentiment that while simply improving existing approaches might not be sufficient, those approaches — such as strengthening protected areas, improving policy regarding the use and protection of natural resources, working in robust partnership with communities who depend on nature for their survival — should always be the first option. At the same time, a growing minority of the conservation community is exploring new tools, such as those offered by synthetic biology, that could complement, and in some cases even reinforce, existing conservation techniques. Conservation is already an integrative discipline, and the incorporation of new tools into the kit should come as no surprise. However, the synthetic biology toolkit is not just a set of capabilities, but in many cases it modifies organisms to become tools in their own right. In this sense synthetic biology, especially gene drives, challenges agreed concepts of tools, organisms and conservation, and must be given special consideration by conservationists and biologists alike, to chart a path forward.

Unfortunately the potential impact of synthetic biology on conservation is a "wicked problem," with no clear route to a solution and no obvious stopping point ((Rittel & Webber, 1973; Redford, Adams, & Mace, 2013). The use of living modified organisms (LMOs), and their impact on biodiversity, remains a controversial but helpful precedent. The recent Convention on Biological Diversity Ad Hoc Technical Expert Group (AHTEG) Report (Ad Hoc Technical Expert Group on Synthetic Biology, 2017) noted that, beyond the experience gained from LMOs already released into the environment, there was limited direct empirical evidence to date on the benefits or adverse effects on biodiversity resulting from the organisms, components and products of synthetic biology. However, some have argued that in relation to gene drive there are crucial differences compared to LMOs and adapted risk assessments may be needed to evaluate their impacts (Simon, Otto & Engelhard, 2018).

For some, interest in synthetic biology represents a fascination with the new, a misplaced hope in a magic bullet technology that will solve heretofore intractable problems. In this view, where conservation has fallen short it has done so because the application of existing techniques was inadequate to address the nature or scale of the problems. Others in the conservation community believe that if the evidence for the utility of a new technique exists, then it should be used regardless of whether the potential for the old approach has been exhausted. In this view, while any new technology must be approached with caution, given the scale and pace of the biodiversity crisis, it makes sense to continue investigating new approaches, bearing in mind the precautionary principle (Harremoës et al., 2002; EEA, 2013), and using them as soon as they can be shown to be effective and safe and acceptable to local communities.

To date, synthetic biology and conservation have proceeded largely in isolation from each other (Redford et al., 2014). The specialties and the scientists who practice them differ in obvious ways, such as training and scientific practice, but in subtler ways including world views, approaches to uncertainty and risk, and value systems. Despite these differences, there is an

increasing sense that, over coming years, conservation and synthetic biology will converge or, as some people fear, collide. New ways to address seemingly intractable problems with scalable technology also present a host of new and unanticipated challenges. It is well noted that an established and continuous dialogue can minimise the potential harm from synthetic biology products that are being developed for multiple purposes, reduce mutual misunderstanding, and maximise their utility for nature conservation (Redford et al., 2014; Revive & Restore, 2015; Piaggio et al., 2017).

Recalling the blurred lines between synthetic biology and the digital sphere, debate about the use of digital sequence information (DSI) corresponding to the DNA of living organisms continues within the Convention on Biological Diversity (CBD), and its Subsidiary Body on Scientific, Technical and Technological Advice which has convened Ad Hoc Technical Expert Groups on both issues. On the one hand this represents an important mainstream interaction between conservation policy and synthetic biology; on the other, the Convention has not yet been able to decide whether synthetic biology should be classified as a new and emerging issue against the criteria set out in Decision IX/29 on Biosafety to the Convention on Biological Diversity (Sections 2.2.1 &2.2.2), and whether or not digital sequence information would be covered by the existing framework of the Nagoya Protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilisation to the Convention on Biological Diversity (Sections 2.2.4 & 2.3.2). Such challenges perhaps reflect other societal concerns regarding the potential interactions between synthetic biology and conservation, as exemplified by the open letter "A call for conservation with a conscience: no place for gene drives in conservation" (Synbiowatch, 2016). However this does not represent the "public" as a whole, and there are limited studies that have examined the public's understanding and views towards synthetic biology and gene drive (Schmidt et al., 2009; Eden, 2014).

Synthetic biology and conservation indeed have the potential to interact in innumerable ways. Conservation may be improved by adapting the tools and processes of synthetic biology to further develop its own goals, much as conservationists did with classical genetics (DeSalle & Amato, 2004). Invasive species may be controlled with limiting gene drive (Case study 1). Oil spills could be remediated with microbes engineered to digest harmful compounds (Dvořák et al., 2017). Infectious and emerging diseases could be treated or prevented (Case study 4), and genetic diversity restored to where it has been lost (Case study 3). Across all such examples, the critical question asks how might such synthetic biology applications impact biological diversity, as measured not just against the current state of biodiversity but against a potential future in which business as usual is allowed to continue. Some applications of synthetic biology in conservation have been particularly controversial and have drawn a great deal of attention. For example, "de-extinction" – the process of creating an organism/animal that is a member of an extinct species or serves as a proxy that may restore their extinct counterparts' lost ecological value (IUCN SSC, 2016) – has been described as being "a fascinating but dumb idea" because it would divert resources away from saving endangered species and their habitats (Ehrlich & Ehrlich, 2014). On the other hand, certain conservation applications, for instance the engineering of microbes to biosynthesise products sourced from threatened species, such as a medically-valuable molecule found in the blood of horseshoe crabs, are already underway (Maloney, Phelan & Simmons, 2018; see Chapter 6 Case study 8 – Horseshoe Crab).

Box 1.1
An introduction to the central dogma of genetics

Phil Leftwich

DNA to RNA to protein

The central dogma of biology has been a remarkably useful model for understanding DNA (Deoxyribonucleic acid), a complex molecule that carries all of the information necessary to build and maintain an organism. DNA can be read by cellular machinery to encode for RNA and protein, and the

three classes of molecule can be considered interchangeable, and common to all life on Earth. Individuals may pass on this information from parents to offspring over generations, or directly to one another through horizontal gene transfer.

Segments of DNA that encode the information for a specific protein are known as genes, and all organisms within a species share a common set of genes, many of which can differ slightly between individuals, the variations being known as alleles. The combined effect of all these allelic differences can have a major role in an organism's suitability for its environment, and helps to define the biological traits of an individual and the species.

DNA structure

The DNA molecule physically manifests as a double helix, composed of two long strands of polynucleotides that run in parallel while winding around each other to resemble a twisted ladder. Each strand is a long chain of smaller units called nucleotides, which may be one of four organic bases — adenine (A), guanine (G), cytosine (C) and thymine (T). The bases along these two strands link to each other in a specific manner – A will only pair with T on the opposing strand, and C will only pair with G. The double helix holds DNA in its linear structure allowing the storage of information via nucleotide ordering along two coding strands. The structure may also be unwound such that each strand serves as a template to form two new identical molecules when cells divide. Stored information sequences can be passed on to descendant molecules as the two halves separate, and can even be recombined between organisms during reproduction, providing the molecular basis for heredity and variation in offspring.

Gene expression

A gene can be defined as a section of DNA that codes for a particular protein, with the order of nucleotides directing the ordered assembly of amino acids into a protein string. Protein strings fold into three-dimensional structures, which in turn determine the function of the folded protein. The process of directing protein synthesis is known as gene expression, and can occur at all times, or in response to particular environmental cues. Given the vital importance of genes in making all of the proteins that enable an organism to function they make up a surprisingly small proportion of the total genome. The human genome is made up of approximately 21,000 protein-coding genes – but this accounts for less than 2 per cent of the nucleotides in the total genome. Despite this, protein molecules form the basis of all living tissues and play central roles in all biological processes. Examples of proteins include antibodies, enzymes and structural proteins and hormones.

Beyond the gene model

The central dogma and gene model serve as a useful basis for introducing concepts of genetics, but these simplifications hide the complexity of how genomes, genes, gene regulatory processes, trait manifestation and other complex genetic phenomena occur. For a more detailed primer on genetics, see Appendix 1 (www.iucn.org/synbio).

1.3 What is synthetic biology?

All living organisms contain shared fundamental components that serve as an instruction set to determine what organisms look like, what they do, and how they function (Box 1.1). While synthetic biology is evolving so rapidly that no commonly accepted definitions exist (Box 1.2), underlying all definitions is the concept that synthetic biology is the application of engineering principles to these fundamental components of biology. As the field grows, more and more disciplines are becoming aligned with it, making it even more difficult to find a single definition (Shapira, Kwon & Youtie, 2017). This assessment uses the operational definition considered by the CBD AHTEG as a useful starting point for discussions about synthetic biology: "a further development and new dimension of modern biotechnology that combines science, technology and engineering to facilitate and accelerate the understanding, design, redesign, manufacture and/or modification of genetic materials, living organisms and biological systems" (UN CBD, 2017).

Humans have been altering the genetic code of plants and animals for millennia, by selectively breeding individuals with desirable features to reassert and accentuate traits in populations over time and in environments formed by husbandry practices, social systems and ecological drivers. The advent of biotechnology allowed humans to more precisely read and edit the code that governs genetics, allowing genetic information and traits to be usefully modified. This is the basis of genetic engineering, and has allowed researchers to speed up the process of developing new breeds of plants and animals relevant to agriculture and medical research.

More recent advances at the intersection of biotechnology, modern engineering, computation and chemistry have enabled scientists to design and synthesise new sequences of DNA from scratch, supporting the design of cells and organisms that do new things — such as produce biofuels, secrete the precursors of clinical drugs or act as biosensors. Many believe that designing novel DNA to obtain specific functions is the essence of synthetic biology.

Synthetic biology has been enabled and driven by the ability to convert and represent DNA base pairs, codons, amino acids, genes and regulatory elements in a digital form (National Academies of Sciences, Engineering, and Medicine, 2017). Digital sequence information not only enables researchers to view and understand the blueprints of an organism in a computational environment, but opens the door to designing, editing and modelling biological components prior to physically producing and inserting them into a cell or organism. The simulation and testing of biological designs using computer software is an emerging opportunity to evaluate biological interactions across organisms, and potentially even ecosystems, prior to the release of a modified organism, but there remain challenges in accurate modelling of complex systems. More generally, increasing access to public digital sequence information, collections of biological components and computer automation has substantially reduced the time it takes to design new biological components and enabled new actors to participate in synthetic biology (Section 6.6).

The early concepts underpinning synthetic biology surfaced over a century ago (Leduc, 1912), more recently being formalised as the fusion of molecular biology and engineering principles. Today, synthetic biology exists as, and is embodied in, a broad set of tools, processes and disciplines. The tools may include CRISPR-Cas9 reagents that are used to cut and splice DNA, as well as DNA sequencers and DNA design software packages. Significant synthetic biology processes include genome editing, whole genome sequencing and functional screening. The disciplines associated with synthetic biology include systems biology, bioinformatics, molecular biology, microbial ecology and plant virology (Figure 1.2). A feature of synthetic biology is that this diversity of fields, and the borrowing of tools from non-synthetic biology domains, makes the taxonomy of synthetic biology challenging. Specific tools or processes can rarely be said to be uniquely tied to synthetic biology; CRISPR-Cas9 may be used in multiple non-synthetic biology contexts, for example, and the products resulting from the use of a tool or process are not always the intrinsic products of synthetic biology.

Synthetic biology is a convergent branch of biology and engineering that is perhaps better articulated not as a list of tools, processes and fields, but rather the use cases for which they are developed and deployed. These use cases are expanding as interactions between nanotechnology, artificial intelligence, robotics and a myriad of biological innovations yield breakthroughs in smart materials, material structures, energy generation, pollution remediation and more. Synthetic biology is only one of a set of new technologies that is being developed and deployed. There is a constant, fluid, and potentially extremely broad interaction and innovation frontier between this “Fourth Industrial Revolution” and biodiversity (World Economic Forum’s System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018). The Fourth Industrial Revolution refers to the fourth major industrial revolution and is characterised by its “velocity, scope, and systems impact” and the combination of technologies from the physical, digital and biological realms (Schwab, 2016).

When applied to conservation, each application, tool and process derived from the various disciplines of the synthetic biology field should be evaluated on the evidence for the positive and/or negative impacts they are likely to have on any given conservation objective. In all cases, assessments must widely investigate how a synthetic biology approach will influence the entire *plurality* of conservation objectives for all biodiversity impacted. Only then can informed decisions be made. Such assessments would assemble a body of knowledge to guide future decision makers through the broad spectrum of synthetic biology applications, and the considerations that should be made in light of their impact on biodiversity conservation.

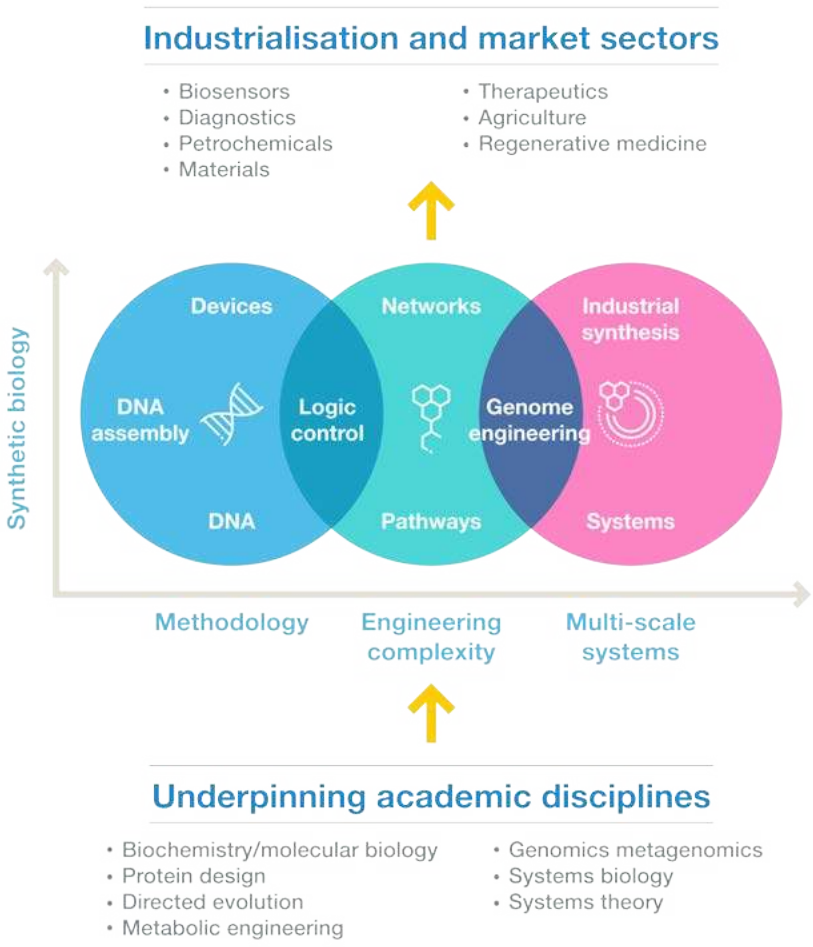


Figure 1.2 What is synthetic biology? Synthetic biology is both a platform technology (building a systematic basis for design—combining biological, engineering, and computational capabilities) and a translational technology (providing the link between a wide range of underpinning disciplines—ranging from biochemistry to systems theory—and practical applications in a wide range of market sectors). Adapted from a figure by the UK Synthetic Biology Roadmap Coordination Group.

Box 1.2
Example definitions of synthetic biology

- A further development and new dimension of modern biotechnology that combines science, technology and engineering to facilitate and accelerate the understanding, design, redesign, manufacture and/or modification of genetic materials, living organisms and biological systems (UN CBD, 2017).
- The application of science, technology and engineering to facilitate and accelerate the design, manufacture and/or modification of genetic materials in living organisms (SCENIHR, SCCS, 2014).
- The deliberate design of biological systems and living organisms using engineering principles (Balmer & Martin, 2008).
- The design and construction of novel artificial biological pathways, organisms and devices or the redesign of existing natural biological systems (The Royal Synthetic Biology Society, 2017).
- The use of computer-assisted, biological engineering to design and construct new synthetic biological parts, devices and systems that do not exist in nature and the redesign of existing biological organisms, particularly from modular parts (International Civil Society Working Group on Synthetic Biology, 2011).
- A new research field within which scientists and engineers seek to modify existing organisms by designing and synthesising artificial genes or proteins, metabolic or developmental pathways and complete biological systems in order to understand the basic molecular mechanisms of biological organisms

and to perform new and useful functions (The European Group on Ethics in Science and New Technologies to the European Commission, 2009).

- A new field defined by the application of engineering principles to living systems for useful applications in health, agriculture, industry and energy (UK BBSRC, 2017).
- A platform technology that enables the design and engineering of biologically-based systems. As a field of science, it encompasses both the biological aspect of designing systems to help understand them, and the engineering aspect of designing systems with the aim of achieving a set endpoint. Thus, overall it involves the design of new living systems that can carry out specific functions or produce products (Parks et al., 2017).
- A new field of research in biotechnology that draws on engineering principles to manipulate DNA in organisms. It allows for the design and construction of new biological parts and the re-design of natural biological systems for useful purposes (OECD, 2016).
- The molecular-biological modification of known organisms which are mostly application-oriented and increasingly based on digital information. These approaches aim at producing chemicals by means of new ways of bio-synthesis or at designing genetic circuits for new sensory and regulatory cell functions in existing organisms. Synthetic biology in the broad sense goes beyond simple approaches for genetically modifying metabolic pathways of organisms (so-called metabolic engineering). For this, computer-assisted design and modelling processes are used increasingly (Sauter et al., 2015).
- An emerging discipline that combines both scientific and engineering approaches to the study and manipulation of biology (NRC, 2013).

1.4 What is gene drive?

In addition to focusing on synthetic biology, IUCN Resolution WCC-2016-Res-086 called for an examination of gene drive systems and biodiversity conservation. Gene drive is a ubiquitous natural phenomenon in which a genetic element improves the chance that it will be inherited at a frequency above the usual 50 per cent by copying itself or selectively eliminating competing elements (Figure 1.3) (Burt and Trivers, 2006; NASEM, 2016a). This potentially allows gene drive elements to spread through populations even without providing a fitness advantage to the individuals carrying the elements, though a fitness disadvantage will slow and perhaps prevent spread. Such spread can be rapid relative to 'normal' gene changes, but still slow relative to genetic elements that can readily transfer between individuals ("horizontal gene transfer") such as viruses or plasmids. Nearly every organism whose genome has been sequenced carries active or broken gene drive elements, which in some species can comprise most of their DNA (Feschotte & Pritham, 2007; de Koning et al., 2011).

Scientists are working to harness gene drive, either repurposing naturally occurring systems or building synthetic versions – engineered gene drives – that might be used to spread engineered changes through wild populations over many generations. Some methods may allow populations to be suppressed by distorting the sex ratio or impairing the fertility of organisms that inherit two copies, which may be relevant for invasive species control. Mathematical models incorporating spatial spread of engineered population suppression gene drives in species such as mosquitoes predict that suppression should not result in extinction absent other ecological pressures (Eckhoff et al., 2017). Many types of gene drive are found in nature; crucially, different mechanisms give rise to different behaviours. Some gene drive elements, including many found in nature and some engineered ones, are predicted to keep spreading to most populations of the target species (Marshall, 2009; Noble et al., 2018). Other types of drive systems are inherently localised due to some form of frequency-dependence; like non-driving genes, engineered local drive systems are not predicted to spread far beyond the populations in which they are introduced (Hoffmann et al., 2011; Marshall & Hay, 2012). For more detailed information on gene drive systems see Appendix 2 (www.iucn.org/synbio).

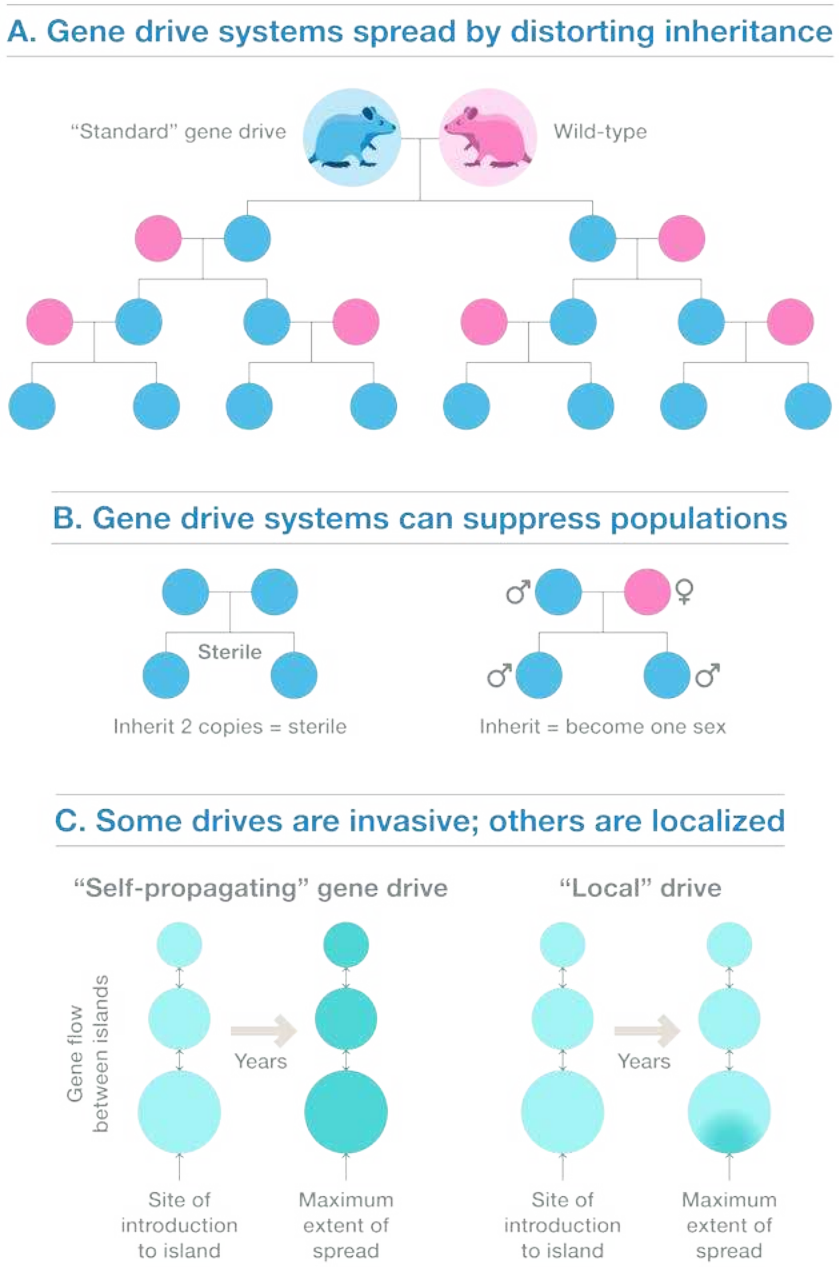


Figure 1.3 What is gene drive? Gene drive systems distort inheritance in their favour, enabling them to spread vertically through populations over generations (a). Some types of engineered gene drive systems can suppress populations, either by ensuring that organisms that inherit one copy from each parent are nonviable or sterile, or by ensuring that organisms inheriting a copy develop exclusively as one sex, e.g. all male (b). Self-propagating gene drive systems are predicted to invade most or all susceptible populations connected by gene flow, whereas the geographic spread of local drive systems is limited by their dependence on the frequency of other genetic elements, reducing their ability to spread or invade populations distant from the release sites (c).

Box 1.3
Modifying epigenomes using synthetic biology

Johanna E. Elsensohn

Epigenetics is a field of study that looks at how environmental (i.e. non-genetic) factors can affect how, whether and when genes are expressed. Epigenetic changes can be transient, present throughout the organism's life cycle, or, in some cases, passed on to subsequent generations. This last possibility, called transgenerational epigenetic inheritance (TEI), is well established in plants, microbes, yeast and nematodes, among other organisms (Rusche, Kirchmaier & Rine, 2003; Casadesús & Low, 2006; Quadrana & Colot, 2016; Minkina & Hunter, 2017). As

epigenetic modifications can target the expression pattern of a specific gene at a specific time, the implications for its use in synthetic biology and engineered gene drive systems could be significant (Jurkowski, Ravichandran & Stepper, 2015; Keung et al., 2015). However, synthetic biologists are only beginning to explore the implications of this research (Rodriguez-Escamilla, Martínez-Núñez & Merino, 2016; Maier, Möhrle & Jeltsch, 2017).

The existence of TEI in mammals remains unclear. First, no mechanisms have been identified, with specific exceptions (e.g. researchers have silenced but not altered the sequences of certain genes of newborn agouti mice by feeding their mothers extra vitamins during pregnancy). Second, mammalian germ cells (that is, eggs and sperm) develop dynamically, which can eliminate epigenetic changes (Feil & Fraga, 2012; Skvortsova, Iovino & Bogdanović, 2018). Challenges to the use of epigenetic modification for conservation or other purposes are similar to those for gene editing, and include a lack of clarity on the stability of engineered epigenetic alterations within and across generations, and the regulations that would apply to the engineered organisms.

Some researchers are exploring the possibility that epigenome therapy may be able to help prime certain genes of threatened species against specific stressors. Epigenome editing has mostly been explored in humans (Kungulovski & Jeltsch, 2016; Holtzman & Gersbach, 2018), but has broader potential (Keung et al., 2015; Sharakhov & Sharakhova, 2015). Such changes would not be passed onto future generations and would not address the underlying problems many species face, but epigenetics may offer a stopgap aid during periods of acute stress, such as drought or increased salinity.

1.5 Values in synthetic biology and biodiversity conservation

Values shape how we individually and collectively assess technologies. Synthetic biology is in that sense no different than other transformational scientific discoveries. Values can be understood as motivational goals deeply embedded in material culture, collective behaviours, traditions and social institutions. They often serve to define and bind groups, organisations and societies (Manfredo et al., 2017). As such, values shape how humans individually and collectively assess new technologies such as synthetic biology. The values underlying public discussion about the use of synthetic biology products are raising a mix of moral, metaphysical, socio-political and ethical questions.

One of the recurring concerns is that synthetic biology interventions are tantamount to "playing God" (Dabrock, 2009; Akin et al., 2017), constituting acts that should not be pursued either because of one's faith-based values, or due to risk of irrevocably perturbing complex natural systems seen to be outside of humanity's control at present. Such values are most apparent, perhaps, regarding issues of species extinction (Sandler, 2012). For synthetic biology and biodiversity conservation, this is particularly relevant for questions regarding creation of proxies for extinct species (IUCN, 2016a; see Section 5.3.2) and the rescue of species facing otherwise intransigent threats (see Sections 5.2 and 5.3.1).

In pursuit of improving human health, a case has been made for putting into place methods that would cause deliberate species extinction – a subject that raises concerns among conservation biologists (Sandler, 2012). Extinction of *Anopheles gambiae* could in theory be seen as a logical endpoint if gene drive approaches for malaria control prove effective (Case study 6). Such deliberate extinction would, however, be unprecedented; despite initial enthusiasm regarding destruction of laboratory stocks of *Variola* smallpox (Arita, 1980), many specialists now concur that retention of these is appropriate (Koplow, 2004; Weinstein, 2011). However, no agency has stated extinction of *Anopheles gambiae* as a goal of suppression gene drive approaches for malaria control (Case study 6), and this would in any case be highly unlikely in the wild (Eckhoff et al., 2017) or in *ex situ* settings, given the number of populations maintained in laboratories around the world (https://www.beiresources.org/MR4Home.aspx).

On the other hand, some researchers and ethicists propose a utilitarian perspective on synthetic biology (Smith, 2013), in which ethical issues surrounding the application of synthetic biology are considered in the light of the potential beneficial outcomes for humanity.

For example, concerning the use of an engineered gene drive to control malaria (Case study 6), ethicists have weighed the moral arguments against modifying a mosquito species with the moral arguments for developing a new tool that could positively impact the caseload of clinical malarial disease (Pugh, 2016; Zoloth, 2016). These utilitarian perspectives also inform concerns about a "slippery slope;" that is, once a certain technology is accepted it may lead to new technologies or new options that would not have been acceptable had they been foreseen at the time of the initial decision (Smith, 2013).

1.6 Size and expansion of synthetic biology funding and markets

Synthetic biology is attracting significant funding from both the public and private sectors. Several reports have tracked investment in synthetic biology. A 2015 report from the Woodrow Wilson Center estimated that US research agencies have invested ~US$ 820 million in public funding (WWC, 2015). Less than 1 per cent of the total US funding was focused on risk research and approximately 1 per cent addresses ethical, legal and social issues (WWC, 2015). Since 2012, the majority of US funding has come from its military funding agencies, which have created multiple programmes around synthetic biology that could have research impacts for conservation (WWC, 2015). For example, the US Defense Advanced Research Projects Agency (DARPA) has developed programmes such as Living Foundries (DARPA, 2018c), Biological Robustness in Complex Settings (DARPA, 2018a), Safe Genes (DARPA, 2018d), Insect Allies (DARPA, 2018b), and in late 2016, issued a call for proposals to develop ecological niche-preference engineering technologies, which would "enable the genetic engineering of an organism's preference for a niche (e.g., temperature, range, food source, and habitat)" in order to lessen their "economic, health, and resource burdens" (DARPA, 2016).

Total European public research funding was estimated at €450 million between 2007–2014 (ERASynBio, 2014). While exact funding amounts are difficult to estimate, China began to invest in public research in synthetic biology through its Ministry of Science and Technology, with additional funding from the National Natural Science Foundation of China and other governmental research and technology programmes starting towards the end of the 2000s (Shapira, Kwon & Youtie, 2017). In 2018 Singapore launched a synthetic biology research and development programme (Ong, 2018). A recent analysis of global markets by BCC

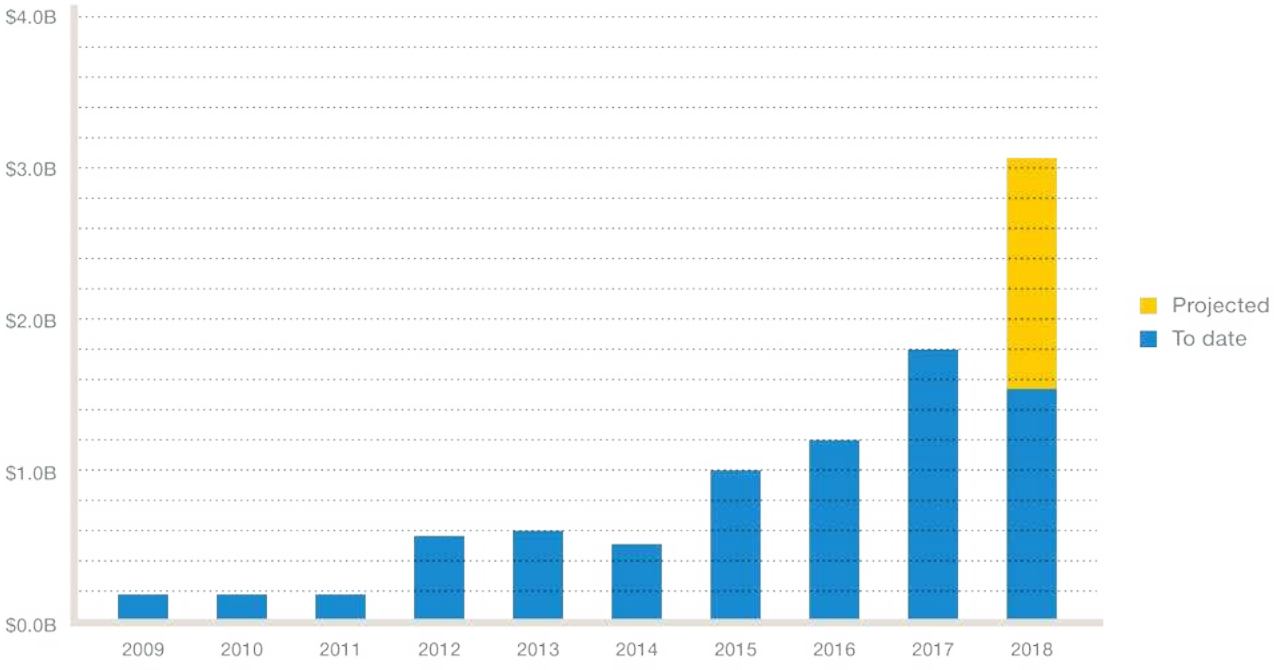


Figure 1.4 Growth in funding for synthetic biology companies. Adapted from Synbiobeta, 2018.

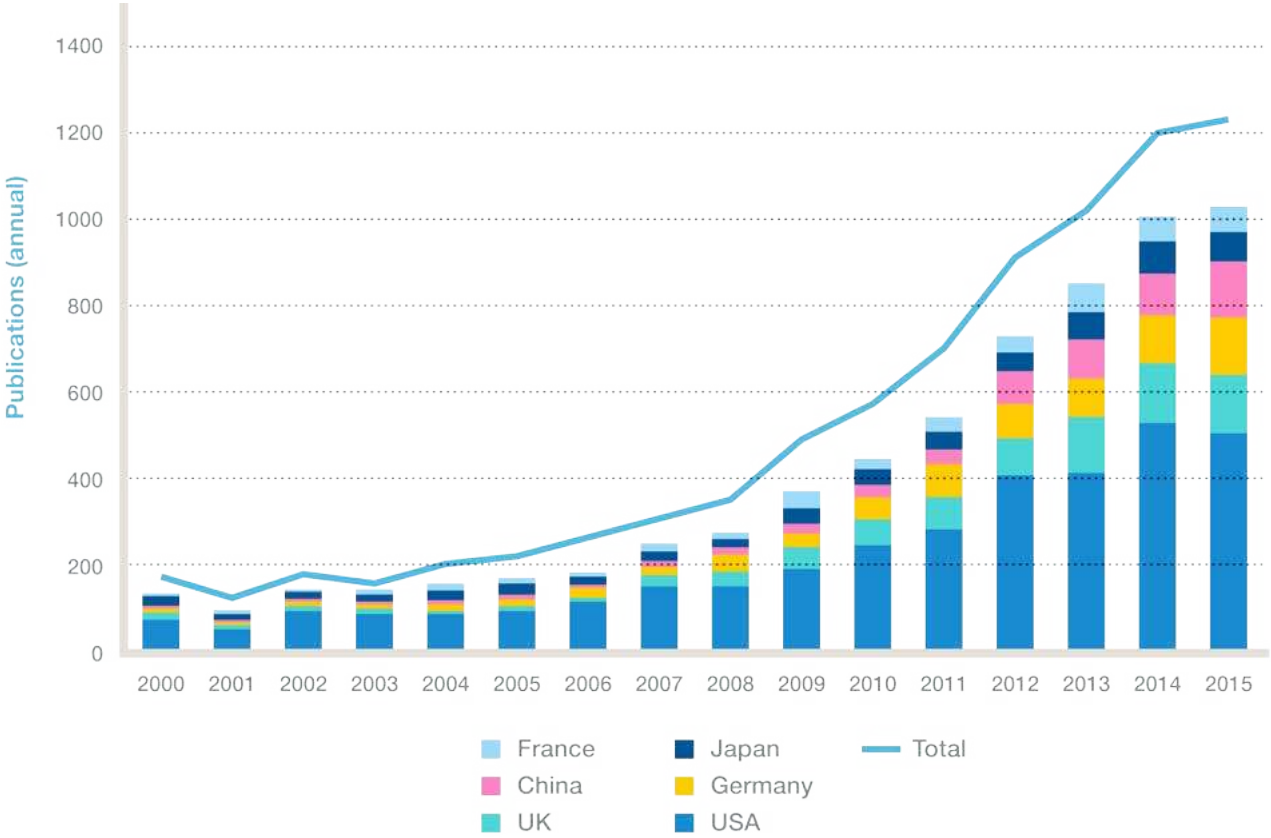


Figure 1.5 Increase in synthetic biology publications. Adapted from Shapira et al., 2017.

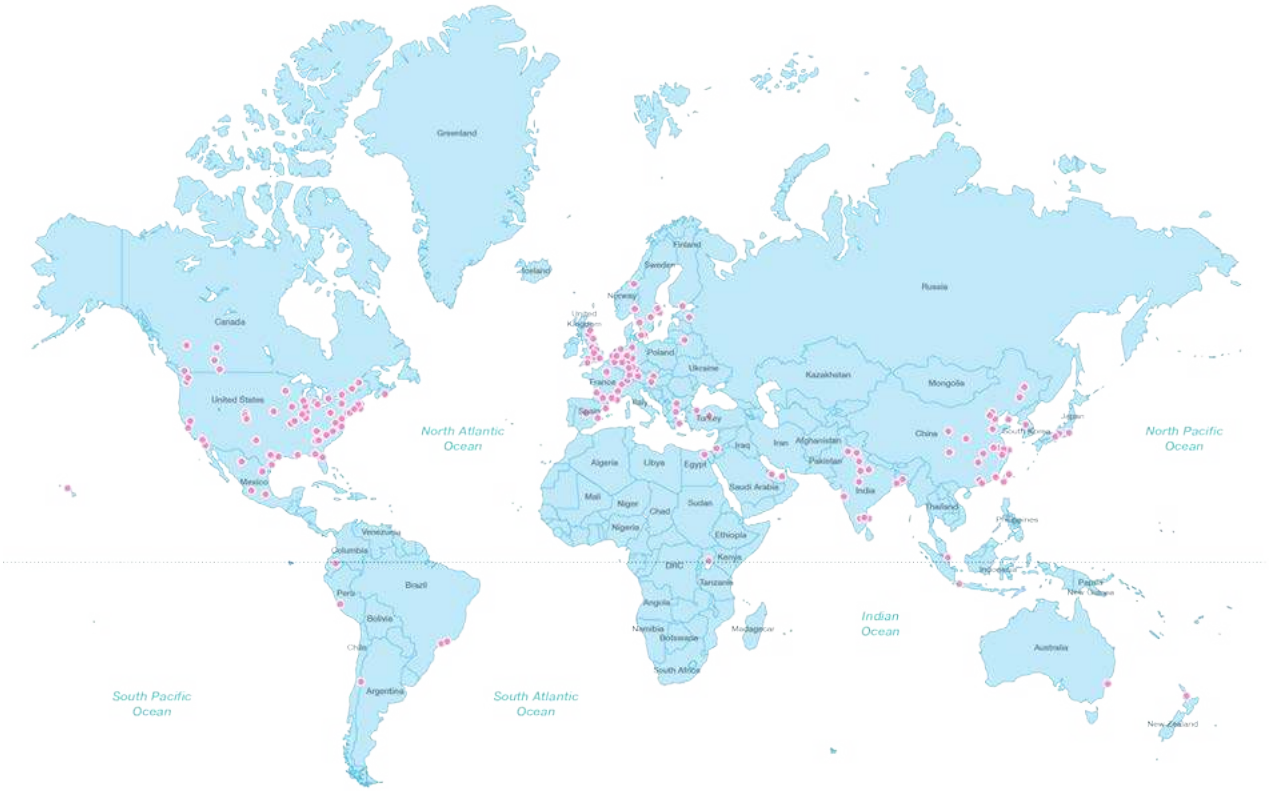


Figure 1.6 2018 iGEM Team Map (iGEM, 2018). The iGEM competition brings together students from universities, high schools and community biotech labs to give them the opportunity to explore synthetic biology. Each dot represents a team or cluster of teams. Multidisciplinary groups work together to design, build, test and measure a system of their own design using interchangeable biological parts and standard molecular biology techniques. Every year nearly 6,000 people participate in iGEM and then come together in the fall to present their work and compete at the annual Jamboree.

Research suggests that in 2017 the global synthetic biology market was valued at US$ 4.4 billion and is expected to grow to US$ 13.9 billion by 2022 (Globe Newswire, 2018). Private investment appears to be growing rapidly. In 2016 over US$ 1 billion was invested in synthetic biology companies, fuelling their rapid growth (Figure 1.4). Figure 1.5 shows the rapid increase in the funding for synthetic biology publications over recent years, and Figure 1.6 shows that the interest in synthetic biology is becoming globally distributed. No data have been traced on the volume of funding for synthetic biology from civil society, including from conservation NGOs, but it is likely relatively small compared to government and industry investment.

1.7 Reports on synthetic biology

Given the funding in synthetic biology it is no surprise that there has been a large number of reports that examine the impacts of synthetic biology and engineered gene drive systems produced by various governments' agencies, think tanks and NGOs (Haseloff Lab, 2018). A sampling of those reports can be found in Table 1.1. These reports represent a broad set of governmental and non-governmental interests and approaches to the field and indicate the extensive consideration that synthetic biology has generated.

Table 1.1 Sample reports examining the impacts of synthetic biology and gene drive systems. For a comprehensive list see Haseloff Lab (2018).

Organisation	Date released	Title	Topics	Reference
Friends of the Earth, Centre for Technology Assessment and ETC Group	2012	Principles for the oversight of Synthetic Biology	Governance, risk assessment	Hoffman, Hanson & Thomas, 2012)
Wildlife Conservation Society	2013	How will Synthetic Biology and conservation shape the future of nature?	Synbio and conservation	(Redford et al., 2013)
European Commission - GEST	2014	Ethics Debates on Synthetic Biology in the Three Regions	Ethics	(Stemerding et al., 2014)
UN Secretariat of the Convention on Biological Diversity	2015	Synthetic Biology - CBD Technical Series No. 82	Risk/benefits	(Scott et al., 2015)
German Office of Technology Assessment	2015	Synthetic Biology - the next phase of biotechnology and genetic engineering	Risk assessment	(Sauter et al., 2015)
UN Secretariat of the Convention on Biological Diversity	2015, 2018	Report of the Ad Hoc Technical Expert Group on Synthetic Biology	Risks/benefits	(Ad Hoc Technical Expert Groups on Synthetic Biology, 2015, 2018)
Dutch National Institution for Health and Environment (RIVM)	2016	Gene drives: Policy Report	Gene drive systems	(Westra et al., 2016)
German Committee of Biological Safety	2016	"Position statement of the ZKBS on the classification of genetic engineering operations for the production and use of higher organisms using recombinant gene drive systems"	Gene drive systems	(GCCBS, 2016)

Organisation	Date released	Title	Topics	Reference
Organisation for Economic Co-operation and Development	2016	OECD Science, Technology and Innovation Outlook	Research trajectories, investment	(OECD, 2016)
US National Academies of Sciences, Engineering, and Medicine	2016	Gene Drives on the Horizon: Advancing Science, Navigating Uncertainty, and Aligning Research with Public Values	Risk assessment of gene drive systems	(NASEM, 2016a)
Australian Academy of Science	2017	Synthetic Gene Drives in Australia: Implications of Emerging Technologies	Risk assessment of gene drive systems	(AAS, 2017)
European Academies of Science Advisory Council	2017	Genome Editing: Scientific opportunities, public interests and policy options in the European Union	Genome editing	(EASAC, 2017)
US Environmental Protection Agency	2017	2017 Update to the Coordinated Framework for the Regulation of Biotechnology	Governance	(US EPA, 2017)
US National Academies of Sciences, Engineering, and Medicine	2017	Preparing for Future Products of Biotechnology	Governance, products, horizon scanning	(NASEM, 2017b)
UN International Treaty on Plant Genetic Resources for Food and Agriculture	2017	Potential implications of new synthetic biology and genomic research trajectories on the International Treaty for Plant Genetic Resources for Food and Agriculture (ITPGRFA or "Treaty")	Digital sequence information	(Welch et al., 2017)
BBSRC-GCRF OpenPlant-Earlham Foundry	2017	Capacity Building for the Bioeconomy in Africa	Capacity building, technology transfer, access	(UK BBSRC, 2017)
African Union	2018	Gene Drives for Malaria Control and Elimination in Africa	Gene drive systems	(NEPAD, 2018)
OECD Working Party on Biotechnology, Nanotechnology and Converging Technologies (BNCT)	2018	Gene editing in an international context: Scientific, economic and social issues across sectors	Gene editing	(Shukla-Jones, Friedrichs & Winickoff, 2018)
US National Academies of Sciences, Engineering, and Medicine	2018	Biodefense in the Age of Synthetic Biology	Biosecurity	(NASEM, 2018)

1.8 International deliberations

It is not just reports that have examined the field. Various international treaties and organisations are currently examining the impacts of synthetic biology and engineered gene drive systems on their respective agreements. Table 2.1 presents these in detail, but in summary they are:

- Convention on Biological Diversity (CBD). Since 2010, the CBD has discussed whether synthetic biology should be classified as a new and emerging issue. An assessment of synthetic biology against the CBD's new and emerging criteria was carried out; however, no definite conclusion was reached. Both the twelfth Conference of the Parties (COP12) and COP13 produced decisions seeking a more robust assessment of synthetic biology against the Convention's new and emerging criteria but this assessment has yet to be completed. Defining synthetic biology as such would officially state that it "needs urgent attention by the Subsidiary Body on Scientific, Technical and Technological Advice" [IX/29 2003], potentially developing new guidance and risk assessments on how synthetic biology and its applications (separate from LMOs) could be utilised in the future by a member state. Decision XII/24 (Secretariat of the Convention on Biological Diversity, 2015) established an Ad Hoc Technical Expert Group on Synthetic Biology that has produced multiple reports and recommendations but which has not yet undertaken the robust assessment against the new and emerging criteria as mandated by the COP (Ad Hoc Technical Expert Groups on Synthetic Biology, 2015, 2018). Current deliberations are also considering whether or not synthetic biology, including engineered gene drive, would fall under the definitions of Living Modified Organisms and thus be subject to the risk assessment requirements of the Cartagena Protocol CBD/SYNBIO/AHTEG/2017/1/3. These deliberations continue.

- Nagoya Protocol. In 2017 the Secretariat of the CBD commissioned a report examining the impacts of digital sequence information as it relates to the Nagoya Protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilization (ABS) to the Convention on Biological Diversity (Wynberg and Laird, 2018). An Ad Hoc Technical Expert Group was also established to provide recommendations for member states on those impacts and a draft decision was submitted with vast disagreements (CBD/SBSTTA/22/CRP.10, 2018). These deliberations continue.

- Food and Agricultural Organization of the United Nations (FAO). In 2017 the International Treaty on Plant Genetic Resources for Food and Agriculture commissioned a report to examine the impacts of synthetic biology and digital sequence information on the Plant Treaty (Welch et al., 2017). These deliberations continue.

- Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). CITES has engaged in a discussion on the question of synthetic products that are indistinguishable from products from listed specimens and the status of modified organisms and products under the convention [Decisions 17.89 to 17.91, 2016; SC69 Doc. 35, 2017].

Similar deliberations have been underway in IUCN, which, through this assessment, has commissioned a broad assessment of the current state of science and policy related to these questions and to identify applications and products that might positively and negatively impact conservation and the sustainable use of biological diversity. As such, this assessment addresses mandates established at the 2016 IUCN World Conservation Congress: "Development of IUCN policy on biodiversity conservation and synthetic biology" (WCC-2016-Res-086), which calls on the Director General and IUCN Commissions to:

examine *the organisms, components and products resulting from synthetic biology techniques and the impacts of their production and use, which may be beneficial or detrimental to the conservation and sustainable use of biological*

diversity and associated social, economic, cultural and ethical considerations; and to

assess the implications *of Gene Drives and related techniques and their potential impacts on the conservation and sustainable use of biological diversity as well as equitable sharing of benefits arising from genetic resources;*

Figure 1.7 situates these mandates within the broader context of IUCN. The Union's membership of governments and non-governmental and indigenous peoples' organisations approved the Resolution, triggering this assessment process. The delivery of the first four operative paragraphs of the Resolution falls under the mandate of the IUCN Commissions and Director General. This report seeks to deliver the assessment elements of the first two operative paragraphs; it has been supported by resource mobilisation (Acknowledgements), and will be finalised based on peer review (Section 3.4.6). Further to the completion of the assessment, the other mandates from the Resolution will be addressed through the development of a draft IUCN policy on synthetic biology and biodiversity conservation, under the mandate of the IUCN Council (Section 7.2). Ultimately, the success of the process should be measured by the uptake of both the assessment and resulting policy across society at large.

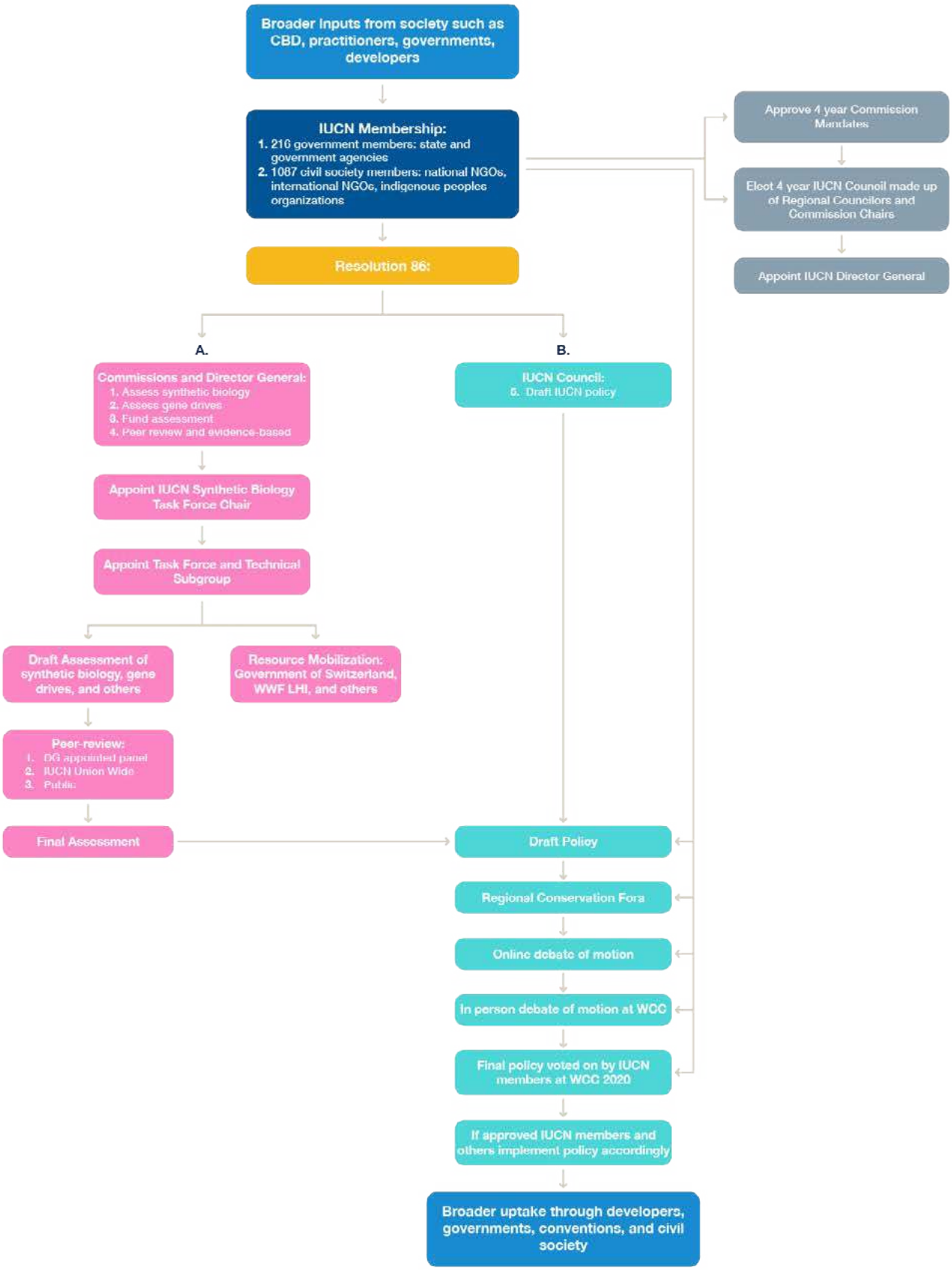


Figure 1.7 IUCN process for developing a policy on synthetic biology and biodiversity conservation.

2. Governance of synthetic biology and biodiversity conservation

Lydia Slobodian, Gerd Winter, Delphine Thizy, Maria Julia Oliva, Ann Kingiri, Adam Kokotovich, Jason Delborne

Image by: Diyana Dimitrova / Shutterstock.com

Understanding the potential implications of synthetic biology for conservation and sustainable use of biological diversity entails examination of the existing governance frameworks applicable to this area, as well as the special governance challenges raised by synthetic biology, including engineered gene drive systems.

This chapter first describes principles relevant to governance of synthetic biology. It then assesses existing governance frameworks and tools applicable to synthetic biology, including international and national law, indigenous, customary and religious governance, and governance by industry and communities of practice. Finally, it discusses challenges raised by synthetic biology, including challenges associated with synthetic biology techniques and practices as well as challenges in engaging with different communities and perspectives.

2.1 Principles

This section highlights principles relevant to the governance of synthetic biology that have featured in the discourse: the precautionary principle; the principle of state sovereignty and state responsibility; principles of access to information, participation and access to justice in decision making; principles associated with indigenous peoples' rights to self-determination and free prior informed consent; and principles of inclusivity and non-discrimination. This is not an exhaustive list of principles, but a selection of principles that appear frequently in ongoing governance discussions on synthetic biology.

2.1.1 Precautionary principle/approach

Scientific uncertainty is a persistent characteristic of environmental governance. The precautionary principle or approach provides a tool for addressing uncertainty in decision making (Wiener & Rogers, 2002; Peterson, 2006). As formulated in the Rio Declaration on Environment and Development, it states:

Where there are threats of serious or irreversible damage, lack of full scientific certainty shall not be used as a reason for postponing cost-effective measures to prevent environmental degradation [Rio Declaration, Principle 15].

This has been reformulated in the preamble of the Convention on Biological Diversity, which reads:

Where there is a threat of significant reduction or loss of biological diversity, lack of full scientific certainty should not be used as a reason for postponing measures to avoid or minimize such a threat.

The CBD has been ratified by 196 states, with the exception of the United States (Table 2.1). Precaution has been referenced in the preamble of the Cartagena Protocol and applied in the articles relating to decision-making procedures. CBD COP Decision XI/11 explicitly applies the approach to synthetic biology, stating:

Recognizing the development of technologies associated with synthetic life, cells or genomes, and the scientific uncertainties of their potential impact on the conservation and sustainable use of biological diversity, urges Parties and invites other Governments to take a precautionary approach, in accordance with the preamble of the Convention and with Article 14, when addressing threats of significant reduction or loss of biological diversity posed by organisms, components and products resulting from synthetic biology, in accordance with domestic legislation and other relevant international obligations [CBD Decision XI/11 para. 4].

In November 2018, the CBD COP further called upon Parties to apply a precautionary approach with regard to engineered gene drives [COP/14/L.31].

Precaution as a legal requirement is multiform and controversial (Marchant, 2003). It has been incorporated into international instruments as well as national constitutions and laws (Fisher, Jones and von Schomberg, 2006; Hanson, 2014). The European Union (EU), for example, has elaborated guidelines on application of the precautionary principle which include a preliminary evaluation of risks and uncertainties to determine when the principle is triggered [EU, 2000]. Other countries, like the United States, have not explicitly included the precautionary principle in their legal system and have

resisted codification of the principle in international treaties, though in practice they may still have adopted measures to manage risk in the context of uncertainty (Hammitt et al., 2005; Hanson, 2014).

While the principle has not yet achieved the status of an international customary rule, it is accepted as an "approach" that guides the interpretation of existing treaty or customary rules (Birnie, Boyle & Redgwell, 2009, p. 163). Whether as a binding principle or approach, there is wide agreement that precaution includes the following core elements (Wiener, 2018, p. 179):

1. a threat of serious or irreversible or catastrophic risk or damage;
2. a stance on knowledge, providing that scientific uncertainty about such risks does not preclude policy measures;
3. a stance on timing, favouring earlier measures to anticipate and prevent the risk;
4. a stance on stringency, favouring greater protection (such as prevention or burden-shifting that prohibits risky activities until they are shown to be safe or acceptable); and
5. a qualifying stance on the impacts of the precautionary measures themselves, calling for them to be cost-effective or weigh costs and benefits, and to be provisional and hence involve reassessment and improvement over time as knowledge is gained (Wiener, 2018, p. 179).

As detailed in Chapters 5 and 6, applications of synthetic biology carry risk that is uncertain and potentially irreversible, making the precautionary principle or approach applicable. There is no consensus on what this means in terms of regulatory measures. Some proponents of synthetic biology claim that some or all of the new techniques should be exempted from current genetically modified organism (GMO) regulation, while others insist that all techniques should be covered by administrative oversight, which may allow for some simplified procedures (ENSSER, 2017). Some civil society and scientific organisations have argued that the precautionary principle or approach necessitates a "moratorium on the release and commercial use of synthetic organisms, cells, or genomes until government bodies, with full participation of the public" have conducted assessments and developed international oversight mechanisms (Friends of Earth (FOE), 2012; https://genedrivenetwork.org/open-letter; http://www.etcgroup.org/content/over-200-global-food-movement-leaders-and-organizations-reject-gene-driveshttp://www.etcgroup.org/content/over-200-global-food-movement-leaders-and-organizations-reject-gene-drives). Others claim that a moratorium on synthetic biology could cripple the field and block potentially beneficial advances, while a more nuanced interpretation of the principle that allows for some, well-regulated risk, could help manage the tension between a desire for caution regarding the risk of intervention and worry about the risks of non-intervention (Wareham & Nardini, 2015).

2.1.2 State sovereignty and state responsibility for international harm

A basic principle of international law is that states have sovereignty over natural resources in their territory as well as responsibility for activities within their jurisdiction or control that cause damage to the environment of other states or areas beyond the limits of national jurisdiction [Stockholm Declaration 1972, Principle 21]. State sovereignty provides the basis for states to make decisions regarding genetic resources and biological diversity within their territory. This includes decisions regarding access to genetic resources that states may subject to requirements for permits and benefit-sharing contracts or make freely available for access and utilisation (Section 2.2.4). State sovereignty also includes decisions relating to activities affecting natural resources in their territory, including decisions on introduction of modified organisms into the environment (Section 2.2.1). Many fora are working on regional and even global harmonisation of state-based standards for risk assessment and management (Tung, 2014).It has been argued, though, that a plurality of approaches may be more realistic and even preferable (Winter, 2016a).

States also have responsibility for transboundary harm. There is an international customary rule that a state must prevent and provide compensation for damage wrongfully caused from its territory to other states [ICJ Pulp Mills 2010]. The International Law Commission has

concretised the general rule by developing Draft Articles on Responsibility of States for Internationally Wrongful Acts, which provide an obligation to make reparation for "any damage, whether material or moral, caused by the internationally wrongful act of a State"[ILC Draft Articles 2001, art. 31].The obligation has been partly applied to biosafety issues by the Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress, which had only 42 Parties as of 2018.

In addition to the "ex post" liability approach, the principle of state responsibility for transboundary harm implicates an "ex ante" approach in the form of a responsibility to conduct environmental impact assessments where there is potential for significant transboundary adverse impact [ICJ Pulp Mills 2010; UNCLOS art. 206]. Depending on scope, this could apply in cases where synthetic biology or engineered gene drives cross boundaries. The Cartagena Protocol further stipulates that export of GMOs requires prior informed consent of the importing state. However, as of 2018, some of the most active states in biotechnology are not among the 171 Contracting Parties of the Protocol, including the United States, Australia, Canada, Russia, Israel and Chile. Failure to comply with prior informed consent and EIA obligations would possibly qualify as a wrongful act in the sense of the international customary rule and Draft Articles described above.

Recognising the potential for harm in the absence of wrongful activities, the International Law Commission of the United Nations developed Draft Principles on the Allocation of Loss in the Case of Transboundary Harm Arising out of Hazardous Activities [2006], which would require states to impose strict liability on operators of hazardous activities, and require operators to have financial security, such as insurance, to cover compensation claims [ILC Draft Principles 2006]. It is however open to debate whether synthetic biology could be considered a "hazardous activity" as understood by the Draft Principles (see Section 2.2).

2.1.3 Access to information, public participation and access to justice in environmental matters

Procedural norms of good governance apply to decision making on activities related to or potentially impacting biodiversity and the natural environment. These include three key components: access to information; public participation in decision-making processes; and access to justice [SDG 16; Rio Declaration Principle 10]. These components have a long tradition in several legal systems, including the United States (Stewart, 2003). They were further elaborated in the Aarhus Convention on Access to Information, Public Participation in Decision-making and Access to Justice in Environmental Matters [1998]. The Aarhus Convention, while European in scope, provides guidance on interpretation of the three aspects, that have been recognised as globally relevant (Morgera, 2005). According to the Aarhus Convention, the principle of access to information requires that any person has the right of access to environmental information held by public authorities, including private actors with public functions, notwithstanding exceptions concerning the protection of privacy, trade secrets and certain public interests [Aarhus art. 4].The principle of public participation provides for a right of the public at large and particularly concerned persons to participate early in decision-making processes in relation to certain hazardous activities or environment-related plans, programmes and executive regulations [Aarhus arts. 6-8]. The principle of access to justice in environmental matters states that any person – which includes any environmental organisation – who considers their rights violated or interests affected by an environmental decision has access to a court or other independent and impartial review procedure to challenge the substantive and procedural legality of the decision [Aarhus art. 9]. The Aarhus Convention explicitly applies these principles to matters related to genetically modified organisms [Aarhus art. 2(3)(a), art. 6(11)].

2.1.4 Peoples' rights to self-determination and free prior and informed consent

Synthetic biology decision making can implicate rights of indigenous peoples and local communities in relation to natural resources and culture. The principle of self-determination of peoples, recognised in the Charter of the United Nations, the International Covenant on Civil and Political Rights, the International Covenant on Economic, Social and Cultural Rights, entails a right to control over natural wealth and resources [UN

Charter art. 55; ICCPR art. 1; ICESCR art. 1]. The UN Declaration on the Rights of Indigenous Peoples and International Labour Organization (ILO) Convention 169 elaborate the rights of indigenous and tribal peoples to participate in the use, management and conservation of resources pertaining to their lands. ILO Convention 169 requires governments to "respect the special importance for the cultures and spiritual values of the peoples concerned of their relationship with the lands or territories, or both as applicable, which they occupy or otherwise use..." [ILO Convention 169 art. 14]. A series of international human rights cases have highlighted the special relationship between indigenous peoples and their traditional territory and resources and found that interference with rights of communities related to their natural resources can implicate the human right to culture [e.g. HRC "Lubicon Lake Band" 1984; IACHR "Awas Tingni" 2001; ACHPR "Endorois" 2009].

In practice, these rights are realised through procedural requirements for involvement of communities in decision making. The UN Declaration on Rights of Indigenous Peoples provides that indigenous peoples shall not be relocated from their lands or territories without their free, prior and informed consent [art. 10]. The concept of free prior and informed consent (FPIC) has been extended to apply to any decision making related to activities affecting the territory or natural resources of indigenous peoples or communities. For instance, financial institutions have included FPIC in the Equator Principles, a risk management framework for determining, assessing and managing environmental and social risk in projects (Amalric, 2005). Human Rights Tribunals have found that FPIC entails good faith and culturally appropriate consultation, sufficient sharing of information including environmental and social impact studies in advance of decisions, and appropriate monitoring [IACHR "Saramaka" 2007; ACHPR "Ogoni" 2001; IACHR "Maya" 2004].

Free, prior and informed consent has been largely discussed in the context of conservation for decisions impacting indigenous peoples and local communities. In its recent report, the CBD's Ad Hoc Technical Expert Group on Synthetic Biology noted that "free, prior and informed consent of indigenous peoples and local communities, might be warranted in the development and release of organisms containing engineered gene drives" (Ad Hoc Technical Expert Group on Synthetic Biology, 2017, para. 25). The AHTEG also stated that the development of synthetic biology technologies "should be accompanied by the full and effective participation of indigenous peoples and local communities" (para. 26). In 2018, the CBD COP called upon Parties and other Governments to obtain, as appropriate, free, prior and informed consent or approval and involvement of potentially affected indigenous peoples and local communities as a prerequisite to introducing engineered gene drives into the environment, in accordance with national circumstances and legislation [COP/14/L.31 para. 9, 11].

2.1.5 Inter-generational equity and sustainable development

Synthetic biology has potential benefits and adverse effects that could affect resource management and economic development now and for future generations. The concept of sustainable development is defined as development that "meets the needs of the present without compromising the ability of future generations to meet their own needs" (World Commission on Environment and Development, 1987). It recognises that economic and social development and environmental conservation are interdependent [Rio Declaration, Principle 4]. It is linked to the principles of intergenerational equity, which entails an obligation of stewardship of the natural environment for future generations, and intragenerational equity which emphasises the need to meet the basic needs of current generations across circumstances and regions (Brown Weiss, 1993; [ICJ Nuclear Test Case, 1995, Weeramantry dissenting; ICJ Gabcikovo-Nagymaros, 1997, Weeramantry concurring; Minors Oposa, 1993]).

The Sustainable Development Goals (SDGs) adopted in 2015 provide globally agreed upon targets for alleviating poverty, ensuring food security, combating climate change and conserving biological diversity. Certain applications of synthetic biology are intended to provide a means for realising sustainable development goals. For example, applications to address invasive species could contribute to goals related to terrestrial

and marine conservation [SDGs 14 and 15], while applications addressing human disease vectors such as mosquitos support achievement of goals on human health and well-being as well as alleviation of poverty [SDGs 1 and 3]. At the same time, some of the risks associated with synthetic biology could affect attainment of these goals in a different way (see Section 2.2). The potential benefits and risks of synthetic biology are discussed in more detail in Chapters 5 and 6.

2.2 Governance frameworks relevant to synthetic biology impacts on biodiversity

Synthetic biology engages existing normative systems, including legal, customary and industry systems, at the international, regional, national and subnational levels. These include frameworks governing risk assessment and management, liability for harm, intellectual property and ownership, and sharing of benefits. Table 2.1 provides a summary of relevant international legal regimes.

Many of the existing governance frameworks were developed in the context of "traditional" genetic engineering and may have to be revised in order to cope with challenges raised by synthetic biology (Wynberg & Laird, 2018). These challenges are addressed in depth in Section 2.3.

This section first explores international and national legal instruments and approaches in relation to risk assessment, liability, intellectual property, and access and benefit sharing. It then briefly discusses indigenous, customary and religious governance, followed by governance by industry and communities of practice.

Table 2.1 International legal frameworks.

Instrument	Description	Relevance for synthetic biology
Convention on Biological Diversity (CBD) Adopted: 1992 Entered into force: 1993 Parties: 196	Global legal framework addressing conservation, sustainable use and sharing of benefits of biodiversity	Creates obligations for each Party to manage risks associated with living modified organisms that could have a negative impact on biological diversity (art. 8(g)) and framework for access and benefit sharing relating to genetic resources (art. 15).
Cartagena Protocol on Biosafety to the Convention on Biological Diversity (Cartagena Protocol) Adopted: 2000 Entered into force: 2003 Parties:171	Protocol to CBD intended to ensure the "safe transfer, handling and use of living modified organisms resulting from modern biotechnology that may have adverse effects on biological diversity..." (art. 1)	Requires sharing of risk related information between exporting and importing Parties and provides guidelines on methodology for environmental risk assessments and considerations in decision-making.
Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress to the Cartagena Protocol on Biosafety (Supplementary Protocol) Adopted: 2010 Entered into force: 2018 Parties: 42	Supplementary Protocol to Cartagena Protocol intended to provide rules and procedures for liability and redress relating to living modified organisms	Provides for national frameworks requiring response measures and assigning civil liability in event of damage resulting from living modified organisms which find their origin in transboundary movement.
Nagoya Protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilization to the Convention on Biological Diversity (Nagoya Protocol) Adopted: 2010	Protocol to CBD providing international framework for access to genetic resources and sharing of benefits arising from their utilisation	Applies to genetic resources that serve as source material for synthetic biology research. Creates ABS framework based on traceability and transfer of material that could be undermined by use of digital sequence information.

Instrument	Description	Relevance for synthetic biology
Entered into force: 2014 Parties: 105		
International Treaty for Plant Genetic Resources for Food and Agriculture (ITPGRFA) Adopted: 2001 Entered into force: 2004 Parties: 144	International regime recognising sovereign rights over plant genetic resources for food and agriculture, and establishing multilateral system to facilitate access to and sharing of benefits from listed plants	Creates ABS system that could be undermined by new techniques using digital sequence information that enable development of new plant varieties without access to the original genetic material.
Agreement on Trade-related Aspects of Intellectual Property Rights (TRIPS) Adopted: 1994 Entered into force: 1995 Parties: 164	WTO Agreement defining obligations to grant and respect patents, including exceptions for patenting of plants, animals and biological processes	Provides forum for ongoing discussions on patentability of genetic resources.
Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) Adopted: 1973 Entered into force: 1975 Parties: 183	Multilateral Environmental Agreement establishing regulations and permitting system covering trade in listed species	Hosts discussions related to 1) synthetic products that substitute or resemble products from a CITES listed species in international trade; and 2) status of genetically modified species and "de-extinction" under CITES.
UN Convention on the Law of the Sea Adopted: 1982 Entered into force: 1994 Parties: 168	Codification of law of the sea including activities and resources in areas beyond national jurisdiction	Provides basis for ongoing negotiation of international agreement on marine biodiversity in areas beyond national jurisdiction, including sharing of benefits from marine genetic resources.
Convention on the Prohibition of Military or Any Other Hostile Use of Environmental Modification Techniques (ENMOD) Adopted: 1976 Entered into force: 1978 Parties: 78	Multilateral instrument prohibiting use of military or hostile environmental modification techniques having widespread, long-lasting or severe effects	Potentially applies to military use of synthetic biology techniques with potential to significantly modify ecosystems.

2.2.1 Risk assessment and regulation

Most countries have national regulatory frameworks for risk assessment and management in relation to genetically modified organisms. The Cartagena Protocol requires Parties to "establish and maintain appropriate mechanisms, measures and strategies to regulate, manage and control risks" connected with the use, handling and transboundary movement of living modified organisms (LMOs), including "possible adverse effects of living modified organisms on the conservation and sustainable use of biological diversity" [Arts. 15, 16]. Where LMOs are intended for introduction into the environment, the decision to allow import must be based on a risk assessment and apply precaution [Arts. 7, 10(6), 15]. Annex III of the Protocol outlines the methodology of risk assessment, including identification of potential adverse effects, evaluation of the likelihood of the effects, evaluation of the consequences of the effects and estimation of overall risk. It also lists points to consider, including the characteristics of the recipient or parental organism, the donor organism, the vector and the insert or modification, as well as a comparison of the unmodified with the modified

Figure 2.1 Countries with national laws on risk assessment and management related to genetically modified organisms. This map shows only those countries whose laws show up in the CBD Biosafety Clearing House or ECOLEX legal database. Lack of inclusion on this map does not mean that the country has no biosafety regulation. Adapted from CBD Biosafety Clearing House; ECOLEX.

recipient or parental organism. National biosafety regulation may provide that certain activities require prior authorisation or notification, containment procedures or other forms of administrative oversight.

The Cartagena Protocol has 171 Parties, but was not ratified by several countries active in biotechnology, as discussed above. Nonetheless, many countries have biosafety legislation in place that fully or partly follows the risk assessment framework outlined in the Protocol. A search of the CBD Biosafety Clearing House and the ECOLEX legal database found 131 countries with national laws on risk assessment and management (Figure 2.1). This includes countries such as the United States, Canada and Argentina that are not Parties to the Protocol.

National risk management legislation applicable to synthetic biology may include a range of legal instruments addressing different sectors and products. In addition to specific biosafety regulations, this may include legislation covering plant-breeding, food and drug safety, pesticides, toxic substances, sanitary and phytosanitary measures, and environmental protection. Some countries may have multiple laws that potentially cover synthetic biology products, as discussed below.

2.2.1.1 Scope of application of regulatory oversight

At COP13 in Mexico in 2016, the CBD Parties noted that it is not clear whether or not some organisms of synthetic biology would fall under the definition of LMO under the Cartagena Protocol [COP13 Decision 17, para. 7]. They stated that the Cartagena Protocol and existing biosafety frameworks provide a starting point for addressing synthetic biology but may need to be updated and adapted for current and future developments and applications, and directed the Synthetic Biology AHTEG to continue deliberating on the matter [COP13 Decision 17, para. 6]. In 2017, the AHTEG concluded that “most living organisms already developed or currently under research and development through techniques of synthetic biology, including

organisms containing engineered gene drives, fell under the definition of LMOs as per the Cartagena Protocol" (Ad Hoc Technical Expert Group on Synthetic Biology, 2017, para. 28). In November 2018, CBD COP14 extended the AHTEG, and emphasised the need for case-by-case risk assessments before organisms containing engineered gene drives are considered for release into the environment and recognised that specific guidance on such risk assessment could be useful [COP/14/L.31 para 9(a), 10].

National regulatory regimes take different approaches in addressing scope of applicability. These are often discussed in terms of "product" or "process" approaches. A "product" approach means that oversight is triggered by certain characteristics of products that are considered to pose a risk, no matter by what processes the product was generated, where a "process" approach means that the product that is subject to oversight is defined by the process of its generation. The United States, Argentina, Canada, the Philippines and Bangladesh have been categorised as having product-based approaches, while Brazil, India, China, Bolivia, Australia, Burkina Faso, the EU and New Zealand have been counted as process-based (Ishii & Araki, 2017). In reality, product-based approaches to regulation often rely upon process-based distinctions, and process-based approaches often consider a combination of product and process-based factors. The usefulness of the product/process dichotomy has therefore been questioned (Kuzma, 2016).

The United States applies what is frequently considered a product approach under the Plant Protection Act (PPA), Federal Insecticide, Fungicide and Rodenticide Act, the Federal Food, Drug and Cosmetics Act, and the Toxic Substances Control Act (Bergeson et al., 2015). However, in some cases agencies may consider process in their decision making. For example, applications for permits for introduction of genetically modified plant pests require a "detailed description of the molecular biology of the system (e.g., donor-recipient-vector) which is or will be used to produce the regulated article" [US 7 CFR 340.4] (Kuzma, 2016). The Toxic Substances Control Act applies to genetically modified micro-organisms defined as "intergeneric" but not physically or chemically mutagenised micro-organisms (Wozniak et al., 2013). Likewise, the Food and Drug Administration (FDA) regulates genetically engineered animals under the "new animal drug" provisions of the Federal Food, Drug and Cosmetics Act, considering manufacturing methods and facilities in its review process (FDA, 2017b). There have been claims that the combination of product and process approaches can open the door for industry to lobby for whichever approach suits their interest. According to Kuzma, "[i]ronically the same GE developers who once claimed that the process of GE does not matter for regulatory purposes are now arguing that changes to the engineering process justify looser regulatory scrutiny" (Kuzma, 2016, p. 166).

Canada likewise bases its regulatory approach on the characteristics of genetically modified products, embedded within its overall framework for regulating "novel products." The trigger for regulatory review of products intended for introduction into the environment is "novelty," whether it derives from genetic modification or other techniques, though the determination of "novelty" may entail process considerations (Montpetit, 2005; McHughen, 2016). For example, the Food and Drug Regulations define "novel food" to include "a food that is derived from a plant, animal or micro-organism that has been genetically modified such that … one or more characteristics of the plant, animal or microorganism no longer fall within the anticipated range for that plant, animal or microorganism" [Canada Food and Drug Regulations B.28.001]. The Seeds Regulations define "Novel Trait" as one that "is not substantially equivalent, in terms of its specific use and safety both for the environment and for human health, to any characteristic of a distinct, stable population of cultivated seed of the same species in Canada" [Seeds Regulations 107(1)].The "substantial equivalence" test has raised criticisms of ambiguity and susceptibility to regulatory capture (Moran, Ries and Castle, 2009). Others have lauded the "novelty" trigger as more practical and scientifically sound than other regulatory approaches (McHughen, 2016).

In contrast, the EU applies what is considered a process approach, under which the process of genetic modification of an organism is the main trigger for oversight. A genetically modified organism (GMO) is

defined as an organism "in which the genetic material has been altered in a way that does not occur naturally by mating and/or natural recombination" (2001/18/EC Art. 2(2)). This definition implies that the application of the recombination technique must result in a changed organism, and hence in a modified product (Callebaut, 2015). Certain techniques are listed as being – among others – genetic modification. They are considered to include not only the transfer of genes between species (transgenesis) but also the reorganisation of genes within a species (cisgenesis) [ECJ Case 528/16, 2018, paras 27–38].

2.2.1.2 Regulatory stages and requirements

Most regulatory systems require prior authorisation for certain types of genetic engineering or release of GMOs into the environment. For activities considered to be low or negligible risk, notification or reporting obligations are used as a form of more lenient oversight. Synthetic biology applications are often subject to step-by-step or staged regulation and monitoring at different levels, from the laboratory to full deployment/release of the organism through potentially other stages such as confined field trials (Figure 2.2). For example, EU Directive 2001/18 sets out a step-by-step approach for introduction of a GMO into the environment, with evaluation of impacts on human health and the environment required at each step. Its preambular consideration 24 explains this as follows:

The introduction of GMOs into the environment should be carried out according to the 'step by step' principle. This means that the containment of GMOs is reduced and the scale of release increased gradually, step by step, but only if evaluation of the earlier steps in terms of protection of human health and the environment indicates that the next step can be taken.

Likewise, in Canada, plants with novel traits, including genetically modified plants, must go through multiple regulatory stages to be approved for environmental release. Stages include, as applicable: import (subject to permit); contained use in a laboratory or greenhouse (subject to biosafety guidelines); confined environmental release (subject to risk management conditions); unconfined environmental release (subject to risk assessment and management and monitoring); variety registration; and commercialisation ([http://www.inspection.gc.ca/plants/plants-with-novel-traits/eng/1300137887237/1300137939635]).

Oversight tools typically distinguish between GMOs made or used in containment and GMOs released to the environment (Prabhu, 2009). For example, Japanese legislation distinguishes between "Type 1 Use" and "Type 2 Use" where "Type 2 Use" describes use where measures are taken to prevent release outside the facility, and "Type 1 Use" refers to all other use where such measures are not taken. Type 1 Use requires ministerial determination that the use will result in no adverse effect if the approved procedures are followed, while Type 2 Use requires confirmation of measures for containment [Japan, Act no. 97 of 2003, arts. 4–15].

In some cases, the areas where the GMO may be released are restricted. In the EU, even if a genetically modified plant was authorised for the EU market, the member states have powers to "opt out" and close areas and even the whole country to its release (Winter, 2016a) [2001/18/EC Art. 26b]. In addition, nature protection, seed protection and other laws may prevent the release of GMOs for specified areas. For instance, in an area under special nature protection the introduction of GMOs may be categorically excluded for reasons of maintaining GM free reference sites, or of preserving the pristine nature. In Germany and

Figure 2.2 Typical stages in risk regulation applicable to synthetic biology.

other states, farmers have agreed to declare regions as to be held GMO-free (GMO Free Europe, 2016).

2.2.1.3 Factors in assessing risks

In assessing risk, national decision makers may be legally required or allowed to take different factors into consideration. Many countries' laws institute administrative bodies and provide them with broadly discretionary powers of oversight [see, e.g. (Saegusa, 1999); Nordrhein-Westfalen Nature Protection Law s. 54]. Other countries' laws set out material yardsticks for oversight in an endeavour to bind administrative decision makers and provide legal certainty for operators [see, e.g. EU Directive 2001/18/EC, Article 4; German Genetic Engineering Act sec. 16]. Commonly, laws provide that impacts on human health and the environment are to be considered.

In addition, some countries include socio-economic concerns as well as impacts on indigenous and local communities. Art. 26 of the Cartagena Protocol states:

The Parties, in reaching a decision on import under this Protocol or under its domestic measures implementing the Protocol, may take into account, consistent with their international obligations, socio-economic considerations arising from the impact of living modified organisms on the conservation and sustainable use of biological diversity, especially with regard to the value of biological diversity to indigenous and local communities.

There are many possible socio-economic considerations that could be relevant to biotechnology regulation, and the ways in which they are taken into account vary across countries (Ludlow, Smyth & Falck-Zepeda, 2014). For example, there are arguments that use of biotechnology can drive change in agricultural practices, and even influence the change of whole regions from sustainable peasant agriculture to industrialised agriculture, as has been observed in Argentina and other countries (Robin, 2010). This type of socio-economic impact could potentially be captured in systems like the EU, which considers impacts on cultivation, management and harvesting techniques.

In some countries, moral values are also considered in risk regulation. Poland, for instance, referred to religious beliefs of its population when prohibiting the cultivation of genetically modified plants, though bringing such plants onto the market was previously authorised by the Commission. The European Court of Justice found the reason not sufficiently substantiated [ECJ Case C-165/08, paras 57–59].

2.2.1.4 Weighing risks against benefits

Many risk assessment frameworks do not allow for analysis of benefits. Some legal systems, including that of the EU, have separate systems for risk assessment – which does not consider costs – and risk management – which can consider regulatory costs and other concerns, depending on the wording of the applicable law (Winter, 2016b).

The United States applies cost-benefit analysis in many, but not all, processes of environmental decision making. In reviewing pesticides, the US Environmental Protection Agency (EPA) considers economic, social and environmental costs to determine whether any adverse effects on the environment are "unreasonable" (NASEM, 2016b). Conversely, in determining safety of food additives, the FDA can only consider whether there is a "reasonable certainty of no harm," and may not take into account other factors (NASEM, 2016b). Cost-benefit analysis has been proposed as an alternative to the precautionary principle as a means for guiding decision makers and ensuring the best outcomes, taking into consideration all possible benefits and risks (Sunstein, 2005).

There are different methodologies for weighing risks and benefits. One example can be found in the EU chemicals regulation [1907/06 "REACH"]. According to Art. 60, an authorisation of marketing of a substance is possible, even if the substance is highly dangerous or considered not to be adequately controlled, "if it is shown that socio-economic benefits outweigh the risk to human health or the environment arising from the use of the substance and if there are no suitable alternative substances or technologies." This is a type of risk-inclined approach, which allows benefits to outweigh any risk, even a serious one. Other systems are risk-averse, allowing only residual risks to be outweighed by benefits (Winter, 2016b).

Those states that allow for the weighing of risks and benefits of products for synthetic biology must consider how to define benefits. The EU chemicals regulation suggests a broad range of concerns including economic interests of suppliers, employment, consumer demand, benefits for human health and the environment, etc. [Annex XVI of the REACH Regulation]. Other approaches would limit benefits to justifiable use values that are expressed in qualitative terms rather than through market prices or survey-based pricing (Ackerman & Heinzerling, 2004; Winter, 2018).

The CBD COP in 2016 invited parties "in accordance with their applicable domestic legislation or national circumstances, to take into account, as appropriate, socio-economic, cultural and ethical considerations when identifying the potential benefits and potential adverse effects of organisms, components and products resulting from synthetic biology techniques in the context of the three objectives of the Convention" [CBD COP13 Decision 7, 2016]. The present study describes certain ways that synthetic biology can be intended to create benefits for biodiversity conservation and sustainable use (Chapter 5) and socio-economic benefits and benefits for human health (Chapter 6), as well as potential negative impacts. For example, the effect of a new product (such as "natural" vanillin produced through synthetic biology) on existing supply chains (such as vanilla supply chains in Madagascar) may have to be weighed against socio-economic benefits of synthetic production (Chapter 6).

Another component of risk-benefit weighing is the testing of alternatives, to determine which could achieve the intended benefit with lowest environmental risks. For example, in evaluating a proposal for modification of a mosquito to eradicate human malaria, decision makers would need to consider alternatives such as vaccination and use of pesticides. Under this concept, it would not be necessary to assess the value of human lives saved and compare them with the loss of biodiversity. It may suffice to examine which of the alternatives – the synthetic biology technique and the application of chemicals – have less harmful impacts on the environment (Winter, 2018).

2.2.1.5 Risk assessment methodologies

The methodology of risk assessment has a common structure throughout national systems, but differs somewhat in terms of depth and width of analysis (Paoletti et al., 2008). One of the most detailed examples is the EU Environmental Risk Assessment methodology (Box 2.1). Most risk assessment methodologies are based on two main components: (1) evaluation of intended and unintended effects, including probability and potential significance of the effects; and (2) comparison of the modified product with existing counterparts (Paoletti et al., 2008). In evaluating potential effects, decision makers can consider information relating to, inter alia, toxicity, persistence and gene transfer, and evaluate potential intended and unintended impacts on target and non-target populations as well as associated social and cultural effects. The comparison of the modified product with counterparts is at the heart of risk assessment. Many countries exempt products from risk assessment where they have a history of safe use. Traditionally the comparison has been between modified and "natural" products, but as genetic modification becomes more common, the definition of "conventional" may change (Paoletti et al., 2008; Pauwels et al., 2013).

Box 2.1

Environmental risk assessment in the EU

The environmental risk assessment (ERA) required by the EU Directive on deliberate release into the environment of genetically modified organisms is defined as "the evaluation of risks to human health and the environment, whether direct or indirect, immediate or delayed, which the deliberate release or the placing on the market of GMOs may pose and carried out in accordance with Annex II" [EU Directive 2001/18/EC, arts. 2(8), 4(2)]. In relation to agricultural plants a Guidance Paper of the European Food Safety Authority (EFSA) distinguishes between seven paths of possible impact (EFSA, 2010):

- Persistence and invasiveness of the GM plant, or its compatible relatives, including

plant-to-plant gene transfer
- Plant-to-microorganism gene transfer
- Interaction of the GM plant with target organisms
- Interaction of the GM plant with non-target organisms
- Impact of the specific cultivation, management and harvesting techniques[2]
- Effects on biogeochemical processes
- Effects on human and animal health

Each specific path must be examined following six steps of ERA [EU Directive 2001/18/EC Annex II sec. C.2].

Figure 2.3 Six steps in the EU environmental risk assessment. Adapted from Directive 2001/18/EC.

Under the EU regulations, different types of information are to be submitted and considered in risk assessment, including information on the molecular and cellular level, the organism and population level, and the ecosystem level, as well as technical information. The information must reveal how the donor organism differs from the recipient organism in terms of functions, reproduction, dissemination, survivability, etc. [EU Directive 2001/18/EC, Annex III].

2.2.1.6 Monitoring

Legislation may provide for monitoring of regulated activities. The United States provides post-market oversight authority to multiple agencies in relation to biotechnology products. The FDA requires reporting from manufacturers and conducts post-market risk assessment and safety inspections in relation to animal drugs, foods and other biotechnology products (NASEM, 2017b). The EPA is required to re-evaluate pesticide products every 15 years, though in practice it has been re-evaluating biotechnology products every 5–6 years. In contrast, genetically engineered organisms that could act as plant-pest can be deregulated upon evidence that they are unlikely to pose a risk, in which case there is little follow-up monitoring or oversight (NASEM, 2017b).

Under EU law, monitoring requirements are different depending on whether a GMO is experimentally released into the environment, or if it is brought to the market with subsequent general release. In the latter case, for instance, the operator is obliged to comply with the authorisation conditions, and in particular with regard to the monitoring scheme, and to continuously report to the competent authority about unexpected

[2] An example for such effects on cultivation practices would include the change of whole regions from sustainable peasant to industrialised agriculture, as has for example been observed in Argentina (Robin, 2010).

incidents during the market placement or release into the environment, be it through case specific or general observations. Likewise, the competent authority is obliged to supervise the monitoring and intervene in case of emergencies [EU Directive 2001/18/EC Article 20]. It has however been found that the monitoring requirements are not well implemented in practice and need to be revised in order to produce more scientifically usable information (Züghart et al., 2011).

2.2.2 Liability

National and international legal systems may provide for liability for environmental damage attributable to synthetic biology. As described in Section 2.1.2, there is an international legal principle of state responsibility for international harm. However, there are few international frameworks that explicitly provide for liability – either on the part of states or on the part of operators – in the context of biosafety. The Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress [Supplementary Protocol] to the Cartagena Protocol provides for states to establish national frameworks for liability in cases of environmental harm caused by living modified organisms. Under the Supplementary Protocol, Parties should require operators to take certain actions in the event of damage, including informing the competent authority, evaluating the damage, and taking reasonable actions to restore affected biodiversity [art. 2, 5]. Where the operator fails to take appropriate response measures, the competent authority may implement such measures and recover from the operator the associated costs. States should also provide for rules and procedures that address damage, including as appropriate, civil liability. Parties may apply existing general rules and procedures on civil liability and/or develop specific civil liability rules and procedures. In either case, under the Protocol they shall, as appropriate address (a) damage; (b) standard of liability (strict or fault-based); (c) channelling of liability; and (d) the right to bring claims. The Supplementary Protocol provides little in the way of binding obligations for civil liability, and has only 42 Parties to date.

European legal instruments apply a principle of strict liability, or no-fault liability, for damage to the environment resulting from certain dangerous activities. The European Convention on Civil Liability for Damage Resulting from Activities Dangerous to the Environment [Lugano Convention] imposes liability on the operator of a dangerous activity for any damage caused by the activity, regardless of fault [art. 6]. Dangerous activities are those which create significant risk for man, the environment or property, and include the production, storage, use disposal or release of genetically modified organisms [art. 2]. The EU Liability Directive applies strict liability to environmental damage caused by a set of listed activities "in order to induce operators to adopt measures and develop practices to minimise the risks of environmental damage so that their exposure to financial liabilities is reduced" [preambular para 2, art. 3(1)(a)]. Listed activities include: "Any contained use, including transport, involving genetically modified micro-organisms" and "Any deliberate release into the environment, transport and placing on the market of genetically modified organisms" [Annex III (10 and 11)].

In Tanzania, the 2009 Biosafety Regulations provide for strict liability in relation to GMOs, including synthetic organisms [§ 3]. The Regulations state:

Any person or his agent who imports, transits, makes contained or confined use of, releases, carries out any activity in relation to GMOs or products thereof or places on the market a GMO shall be strictly liable for any harm, injury or loss caused directly or indirectly by such GMOs or their products or any activity in relation to GMOs [§ 56(1)].

Damage to the environment or biological diversity is explicitly included as a type of harm covered by this provision [§ 56(2)]. In these cases, compensation includes the cost of restoration and the cost of preventive measures, where applicable [§ 56(4); 58]. It also applies to harm or damage caused to "the economy, social or cultural principles, livelihoods, indigenous knowledge systems, or indigenous technologies" [§ 59]. The Regulations require operators to take out a policy of insurance against liability [§ 35(1)].

Harm caused by synthetic biology could lead to civil liability under common law principles of tort, or civil law delict. For example, intrusion of modified organisms onto private property could give rise to

claims of nuisance or trespass (Strauss, 2012). In the United States and Canada, farmers have brought lawsuits against biotechnology companies alleging contamination of their fields with genetically modified crops which rendered their yield less valuable or made it impossible for them to achieve organic accreditation (Rodgers, 2003). To bring a tort suit alleging environmental harm from synthetic biology, claimants would need to show standing, causation and damage, as well as fault or strict liability. Each of these elements could be challenging in the context of synthetic biology. Where damage is to an environmental interest rather than a private person, it may be difficult to prove standing. Some of the potential damage from synthetic biology is extremely attenuated; even where it is possible to show "but for" causation, there may not be a sufficiently close causal link between the activity and the damage to show liability. Fault-based liability may be difficult to prove and ineffective; if significant harm occurs despite best safety practices, the cost may lie with the state. Strict liability is typically reserved for particularly hazardous activities or activities listed in statute, and may not be available for harm caused by synthetic biology in many jurisdictions.

2.2.3 Intellectual property

There are differences in how countries deal with inventions and discoveries linked to genetic resources. These can promote or limit development or use of synthetic biology in conservation. While intellectual property decisions are made mainly at national and regional levels, international law, including bilateral treaties on trade and intellectual property, has played a role, e.g. through the harmonisation of patent and plant variety rights.

In general, industrialised countries allow the patentability of genes and gene sequences (Kumar & Rai, 2007). For example, in 1998, the EU harmonised patent law relating to biotechnological inventions and – though excluding the discovery of a gene or gene sequence from patentability – allowed for an isolated gene or gene sequence to constitute a patentable invention, if it met other patentability criteria. In the US, a recent Supreme Court decision found isolated genomic DNA not to be patentable, based on the law of nature exception to patentability [Association for Molecular Pathology v Myriad Genetics, Inc]. However, the Supreme Court maintained non-naturally occurring molecules may be patented, which may limit the impact of the finding in fields such as synthetic biology (Holman, 2014). Developing countries, for example in Latin America, tend not to allow the possibility of patenting genes and gene sequences (Bergel, 2015). For example, in Brazil, biological material, including the genome or germplasm of living organisms, found in nature or isolated therefrom, is not considered an invention [Industrial Property Law, art. 10].

Intellectual property in organisms, including genetically modified ones, are also treated differently by different states. While the United States provides for patent rights in plants and animals under certain conditions (Rimmer, 2008), the EU allows patenting of microorganisms but excludes patenting of plant and animal varieties [EU Directive 98/44/EC Art. 4; Regulation (EC) 2100/94 Art. 1]. In the EU, intellectual property in plant varieties is only possible in the form of plant variety protection. Farmers are allowed to further propagate their plants and develop new breeds (farmers' and breeders' exemptions) [Regulation (EC) 2100/94 Arts. 13 and 14]. The EU does not provide for intellectual property rights in animals, so that in practice trade secrecy protection is used as a substitute [EU Directive 98/44/EC Art. 4 (1) (1); Winter, 2016]. This means for products of synthetic biology that, for example, the malaria vector mosquito that is engineered to be non-reproductive (Case study 6) would be patentable in the United States but not in the EU; the engineered blight resistant chestnut (Case study 4) would be suitable for patent as well as plant variety protection in the US, but only for plant variety protection in the EU. Modified microorganisms would be patentable in both systems. Methods of plant and animal production are also suitable for patenting. This is however excluded in the EU if the processes are "essentially biological" [EU Directive 98/44/EC Art. 4 (1) (2)].

Proponents of intellectual property protection view it as a tool indispensable to promote innovation in synthetic biology (Calvert, 2012). J. Craig Venter, co-founder of Synthetic Genomics, views intellectual property as fundamental for "a vital and robust science

and biotechnology industry"(Nelson, 2014). Others in the field of synthetic biology worry about negative impacts of intellectual property and advocate for more open innovation, in line with experiences in engineering and computer science. For proponents of open innovation, intellectual property in the context of synthetic biology may create a "perfect storm" (Rai & Boyle, 2007). As in other fields, patents may be both too broad (e.g. foundational patents) and too narrow (e.g. patent thickets) that stifle innovation (Martin, 2008; Winter, 2016b).

Openness in synthetic biology is often adopted also as a fundamental principle – though such principle is not always interpreted in the same way (Calvert, 2012). Several initiatives are promoting the synthetic biology commons. For example, the iGEM Registry of Standard Biological Parts is a growing collection of genetic parts that can be accessed to build synthetic biology devices and systems (Section 6.6). This Registry is an open community with a "Get & Give (& Share)" philosophy. Users get parts, samples, data and tools – and give back the new parts they have made. They also share experiences in the open community.

Commentators have compared these efforts to the open-source software model, as an alternative to proprietary rights (Kumar & Rai, 2006). Unlike software, though, copyright does not apply to synthetic biology products. Moreover, the modularity of synthetic biology makes it difficult to mediate how its parts are shared and re-shared (Pottage & Marris, 2012). As a result, the BioBricks Foundation, created in 2006, has developed tools such as BioBricks Public Agreement and OpenMTA, which facilitate access to synthetic biology parts as a public access resource, but impose no obligation on users to 'return' derivative products to the common pool. This is due, in part, to uncertainties as to the existing ownership status of parts, but also to a recognition that different forms of property may not only coexist in synthetic biology, but also contribute to mutual flourishing (Calvert, 2012; Pottage & Marris, 2012).

In terms of intellectual property rights, synthetic biology has been characterised as a tug-of-war between open and proprietary approaches. It may be that such dichotomy is not so clear, but rather that tools such as the BioBricks Public Agreement and OpenMTA are leading to a "diverse ecology" of both proprietary and open systems (Calvert, 2012; Grewal, 2017). Such a system may see a role for patents, particularly for more complex inventions. As explained in *Nature* through a Lego analogy, "the bricks would be free but a design for a complex rocket ship made of hundreds of Lego pieces would be patentable"(Nelson, 2014).

Intellectual property may also be one of the tools used to safeguard synthetic biology commons. As products of synthetic biology do not have copyright protection, it may be possible to create patent-based commons such as the one established by the group Biological Innovation for Open Society (BIOS). Cost may be a hindering factor (Kumar and Rai, 2006). *Sui generis* intellectual property systems may be developed, such as has been done for plant varieties, databases and – in some countries – traditional knowledge. Contracts may also be used to guarantee access to synthetic biology parts and – possibly after some time – to resulting products.

2.2.4 Access and benefit sharing

The CBD recognises that the sovereign rights of countries over natural resources extend to genetic resources, and access to such resources is subject to national authority and regulation. The Nagoya Protocol to the Convention on Biological Diversity affirms that these sovereign rights entail the right to regulate access to genetic resources and negotiate terms for the fair and equitable sharing of benefits from their utilisation. Both instruments recognise rights of holders of traditional knowledge associated with genetic resources to provide approval for and be involved in utilisation of such knowledge and to share in resulting benefits. These provisions are relevant to synthetic biology insofar as it is based on genetic resources accessed for their utilisation (UN CBD, 2015).

Under the Nagoya Protocol, access to genetic resources should be based on prior informed consent and mutually agreed terms, subject to legislative

and regulatory requirements established by the countries where these resources are accessed. Many countries, including, for instance, the UK and Germany have decided not to introduce restrictions on access to their own resources, though, as described below, these countries have requirements on compliance with access rules in other countries. An increasing number of countries, however, have established national frameworks to regulate access to genetic resources within their territories.

Ownership of genetic resources is defined through national laws and regulations. Most countries that have introduced national frameworks for access and benefit sharing distinguish between biological resources, generally owned by private or public persons, and genetic resources, generally owned by the state [absch.cbd.int]. In some countries, such as in South Africa, the state is a trustee of biodiversity, but it does not have ownership over genetic resources, unless these resources occur in public land [South African National Environmental Management Biodiversity Act, 2004]. The landowner or local communities in South Africa own both the biological and genetic resources on their property. Nevertheless, bioprospecting in South Africa requires not only prior informed consent from the owner of the land where plant material is collected, but also the competent authorities, and benefits arising from utilisation of genetic resources are channelled through the state [South African National Environmental Management Biodiversity Act, 2004, art. 3, 81, 85]. In both cases, access to genetic resources is predicated on permits from competent authorities and agreements for sharing of benefits. These requirements would apply to genetic resources accessed for the purpose of synthetic biology.

The Nagoya Protocol aims at ensuring compliance with provider state requirements through corresponding user state obligations. User states are obligated to take "appropriate, effective and proportionate legislative, administrative or policy measures" to ensure that researchers utilising genetic resources within their jurisdiction have accessed them in accordance with the provider state requirements [art 15]. Such requirements also apply to synthetic biology involving genetic resources obtained from a provider state.

Disclosure requirements in patent law provide a mechanism for ensuring compliance with ABS regulations, by requiring patent applicants to disclose the origin of genetic resources on which the invention was based, facilitating confirmation that ABS procedures were followed. A 2017 study published by the World Intellectual Property Organization found that over 30 countries have established specific disclosure requirements related to genetic resources and/or traditional knowledge for patent applications (WIPO, 2017). For example, Article 26 of the Chinese Patent Law requires that the applicant for a patent on an invention-creation accomplished by relying on genetic resources indicate the direct and original source of the genetic resources. Under the Chinese Patent Law, patent rights may not be granted for inventions that are accomplished by relying on genetic resources that are obtained or used in violation of the provisions of laws and administrative regulations [Chinese Patent Law, art. 26].

There is an ongoing negotiation on a new international agreement on marine biodiversity in areas beyond national jurisdiction, including questions of sharing of benefits from genetic resources originating in the high seas or the deep seabed [UNGA Res. 72/249, 2017]. The implications of synthetic biology and associated tools such as digital sequence information have become part of the discussion.

Synthetic biology tools such as digital sequence information challenge ABS frameworks by impeding traceability, as discussed in Section 2.3.4. There are also questions of how to address benefit-sharing questions where inventions involve genetic elements from multiple organisms including organisms both within and beyond national jurisdiction, elements which are functionally identical in different organisms, and elements which are used in the research process but not found in the resulting invention (Bagley & Rai, 2013; Bagley, 2016). The global ABS mechanism is based on the premise that benefit sharing is an important incentive and source of funding for conservation. The challenges raised by synthetic biology could impact this intended contribution to conservation and sustainable use.

2.2.5 Indigenous, customary and religious frameworks

Statutory frameworks are not the only sources of law relevant for synthetic biology. Legally binding norms and authorities governing research and use of synthetic biology can derive from religious, indigenous or customary systems. Multiple legal and normative systems may overlap in the same geographical space, community or subject field (Figure 2.4; Meinzen-Dick & Pradhan, 2002). This legal pluralism is important for synthetic biology, as researchers, regulators and users of synthetic biology may be faced with a maze of legal rules from different sources. Failure to navigate these rules can result in violations that lead to conflict.

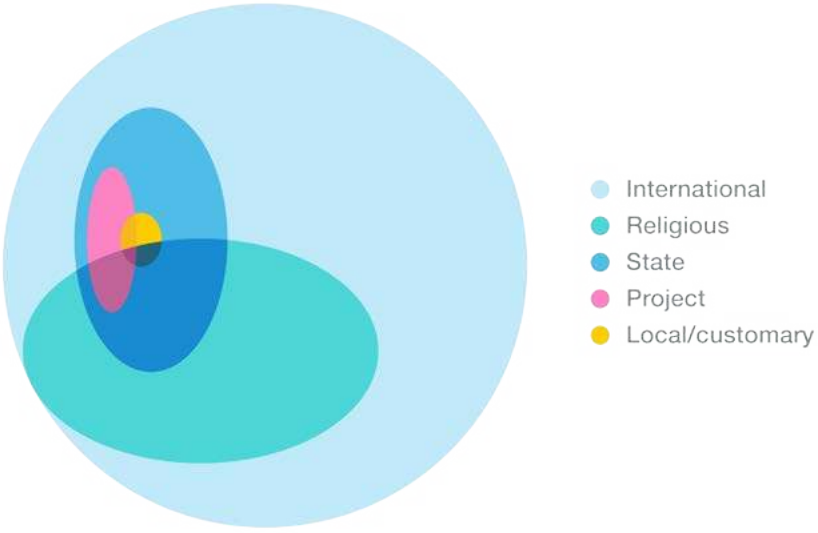


Figure 2.4 Overlaps in normative systems. Adapted from Meinzen Dick and Pradhan, 2002.

Many countries formally recognise indigenous, customary or religious law as well as civil and common law in national legal systems. An IUCN analysis in 2011 found that 60 per cent of the world's countries have constitutional provisions relevant to customary law, ranging from provisions that protect cultural practices to provisions that define customary law and its legal weight (Cuskelly, 2011). In other countries, legal principles or norms from customary or religious systems can be incorporated into legislation. Indigenous or religious authorities can be legally granted exclusive or shared jurisdiction over specific territory or subject matter, or granted the right to participate in national decision making (Cuskelly, 2011). Even where non-statutory law is not formally recognised, it has legal weight within the communities and territories where it is practiced.

The CBD AHTEG has noted that customary law of indigenous peoples and local communities should be taken into account in implementing risk management measures for synthetic biology [CBD/SBSTTA/22/4, 2018, para. 47]. However, there have been few analyses of application of indigenous or customary law to synthetic biology or genetic engineering more broadly. Some of the most advanced research addresses Maori perspectives of synthetic biology and its products and processes. A recent report explored how moving genes between species, introduction of genes from non-native species, extraction of genetic material from an organism and other practices associated with synthetic biology would have direct implications for Maori values, concluding that there are differing positions and interpretations, and that the perceived potential benefits of the technology may vary according to the intended use of the techniques (Mead, Hudson & Chagne, 2017).

Several groups of indigenous peoples have developed formal statements and declarations on the topic of genetic technologies. Many of these assert the right to free, prior and informed consent for research relating to their biological resources, and restrict patenting of such resources (Mead & Ratuva, 2007). The Statement

of Bioethics Consultation from the Tonga National Council of Churches establishes the principle that "scientific and commercial advances should not be allowed to proceed past the deliberations necessary to provide for their social, moral and ethical control" (Tonga National Council of Churches, 2001).

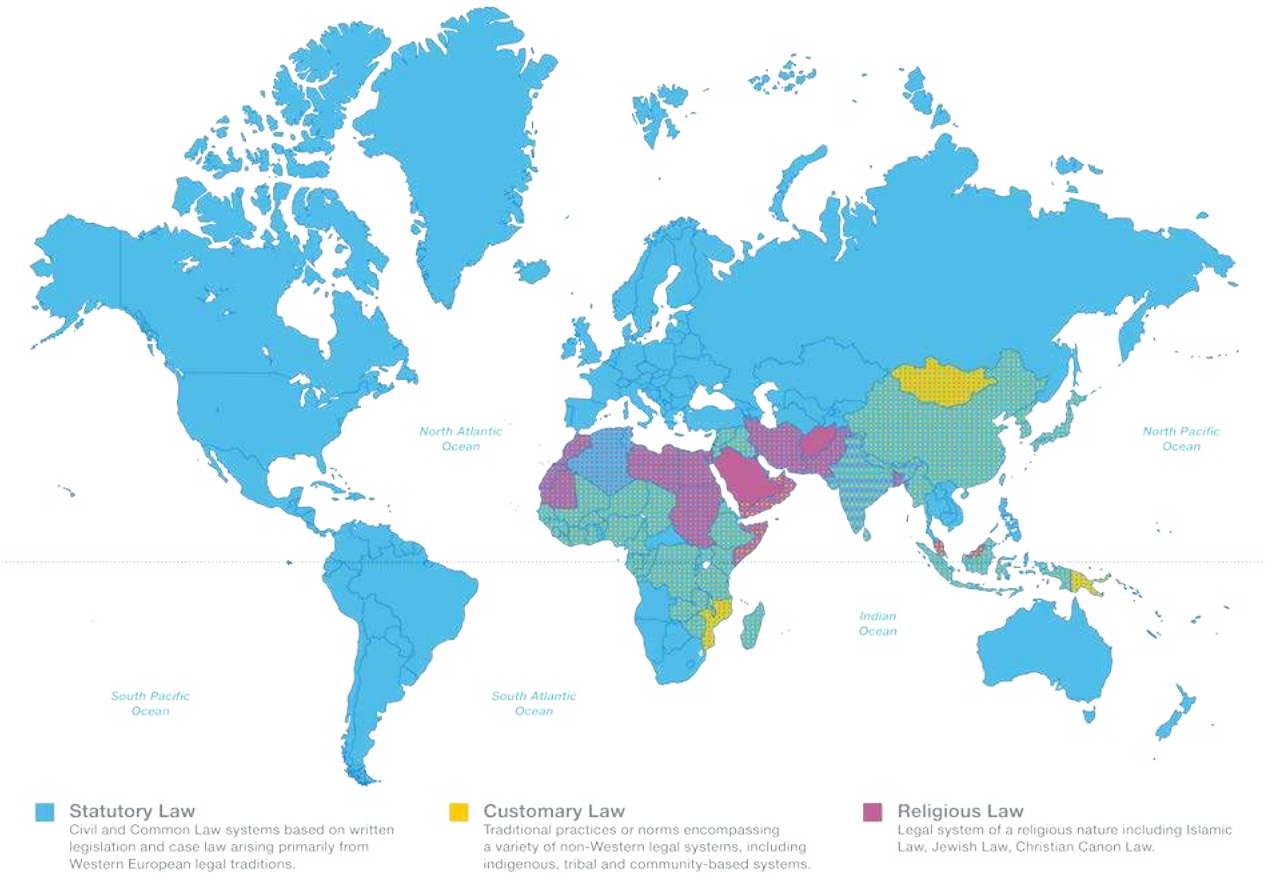


Figure 2.5 World legal systems. Adapted from a map by the University of Ottawa.

There has been some examination of the interaction between customary law and intellectual property aspects of biotechnology. While traditional knowledge is legally protected under the Nagoya Protocol, in practice legal frameworks for ABS and patenting of genetic material focus on statutory law and may exclude customary legal systems relating to property rights and the status of genetic resources (Vermeylen, 2010).

Synthetic biology has spurred active discussion by religious legal experts, raising questions ranging from whether modern biotechnology amounts to "playing God" to whether laboratory meat can be considered kosher or halal (Dabrock, 2009; Gross, 2014). While these discussions influence ethical perspectives on synthetic biology, as discussed in Section 2.3.9, they also relate to applicability of religious law to synthetic biology and constitute a form of governance separate from the role they play in influencing governance under statutory structures. In his 2015 *Encyclical*, Laudato Si, Pope Francis called for "a broad, responsible, scientific and social debate" regarding genetic modification, which he characterised as a "complex environmental issue," recognising both the potential benefits and the ethical questions (Francis, 2015). In 2010, the Church of Scotland produced a report finding that "synthetic biology does not put humanity on a par with God," as synthetic biology techniques do not amount to "ex nihilo creation," but should be guided by humankind's special responsibility for the rest of creation under the doctrine of "Imago Dei" (Church of Scotland, 2010). The Catholic Commission of the Bishops' Conferences of the European Union (COMECE) issued an opinion on synthetic biology in 2016, also finding that synthetic biology techniques do not amount to "playing God" and recognising the potential benefits arising from synthetic biology while calling for appropriate governance measures and public participation (COMECE, 2016; Heavey, 2017). These documents do not constitute sources of binding canon law, but

they do provide a sense of how the Catholic system may view synthetic biology activities and products.

Use of synthetic biology implicates religious law particularly in the context of food. Synthetic meat production could reduce land and water use, with positive benefits for conservation, but there are questions as to how such meat would fit into religious dietary systems (Wolinsky & Husted, 2015). Rabbis at Yeshiva University in Israel have argued that, depending on the circumstances, even artificial pig could be kosher, and could be eaten with dairy ([https://www.ynet.co.il/articles/0,7340,L-5185466,00.html]). Cultured meat could potentially also be halal, depending on the origin of the source cells and the medium used (Hamdan et al., 2018).

2.2.6 Governance by industry and communities of practice

Non-state actors can play an important role in regulating new technologies where the technologies develop rapidly, risks and benefits are uncertain, and there is a need for specialised knowledge (Abbot, 2012). In relation to synthetic biology, there is a growing body of standards created and imposed by industry, researchers and communities of practice. The emerging private sector of synthetic biology uses so-called 'soft' standards, which can facilitate norms and behaviour within the sector, and impact how synthetic biology is perceived by the society (Parks et al., 2017). The soft standards applied by the industry are not binding or legally enforced; instead they rely on personal values and are often 'borrowed' from other relevant standards and more established industries, such as biotechnology and genetic engineering.

Scientists working on engineered gene drive applications have had numerous conversations on self-governance and good practices for safe and responsible research. In 2015, prominent engineered gene drive researchers working on different projects published recommendations for safeguards to contained experiments of engineered gene drive (Akbari et al., 2015). There are ongoing attempts to organise a more formal coordination of researchers working on engineered gene drive technology. For example, the Foundation for the National Institutes of Health convenes the Gene Drive Research Consortium to discuss communication, safe testing and engagement in relation to gene drive technology (FINH, 2018a). The safety board of the International Genetically Engineered Machine (iGEM) international student competition has established a policy specifically discussing safety of their projects and developed a separate policy on work related to engineered gene drive systems and how to prevent accidental gene drive release. These guidelines were established after a team of students attempted to reproduce a scientific paper discussing engineered gene drive development, though discussion of an engineered gene drive policy preceded the incident (iGEM, 2017). The do-it-yourself biology community has developed a code of conduct, which generally draws from good practices applied by the scientific community (DIYbio, 2011).

The role of funding organisations is also important for the governance of research. In its report on gene drive the American National Academies of Sciences, Engineering and Medicine recommended several actions to the funders of research, including the need to collaborate with scientists and regulators to "to develop oversight structures to regularly review the state of gene drive science and its potential for misuse" [recommendation 8.7] (NASEM, 2016a). In addition, the Presidential Commission for the Study of Bioethical Issues established the responsibility of funders to promote some key principles for a responsible research and use of synthetic biology (Weiss, Gutmann and Wagner, 2010). In response to these calls, a number of organisations sponsoring or supporting gene drive research have agreed to a set of principles for responsible research (Emerson et al., 2017). Beyond the key principles, this forum of supporters and sponsors holds regular meetings to discuss key issues around gene drive research, including topics like data sharing, regulatory capacity, etc. (FINH, 2018b).

Several academies of sciences have been looking at synthetic biology or engineered gene drive, trying to establish some recommendations for researchers but also beyond this community proposing guidance for regulators, decision-making authorities and more generally the public (Table 1.1).

2.3 Governance challenges raised by synthetic biology and conservation

Synthetic biology challenges existing governance systems in many respects, of which only a few will be addressed here. New techniques of genetic modification and characteristics of novel organisms create questions relating to the applicability of existing regulations and the methodology of risk/benefit assessment. The potential intended and unintended spread of synthetic biology products, including engineered gene drive, raise challenges for mitigation, liability and compensation systems relating to transboundary harm. Tools and practices associated with synthetic biology, such as use of digital sequence information and the growing "Do-it-yourself Biology" (DIYbio) community, potentially undermine enforcement approaches predicated on monitoring, regulating and tracking genetic material and researchers. Different countries may have different levels of capacity to engage in synthetic biology research and provide effective regulatory frameworks and oversight. A multitude of social, ethical and practical concerns also surround synthetic biology, including the question of moral hazard and concern about sources of funding for research. Engaging with these questions and perspectives creates challenges of its own. There may be particular challenges for developing countries related to research and governance capacity.

2.3.1 Applicability of existing regulations to new techniques

There is a debate over whether existing regulations developed to manage genetic engineering are also applicable to new techniques of synthetic biology. This question goes to the heart of concerns that existing legislation is not adequate to address changing genetic technology. Many regulatory systems were developed for the paradigm of transfer of genetic material (DNA, RNA, etc.) between species – transgenesis. Such systems may not apply to mutagenesis – techniques for modifying the genome without introducing foreign DNA (Duensing et al., 2018). Engineered gene drives may fall into an area of regulatory ambiguity, uncertainty or even overlap – it may not be clear how they fit into existing frameworks addressing pest control, animal drugs, toxins or environmental protection (Oye et al., 2014).

As outlined above (see Section 2.2.1), in the EU, the definition of GMOs and thus the scope of regulatory oversight is very broad, but certain techniques are excluded if they "have conventionally been used in a number of applications and have a long safety record" [2001/18/EC]. Mutagenesis was initially classified as one of those techniques [2001/18/EC Art. 3 with Annex I]. In July 2018, the EU Court of Justice decided that while physical and chemical mutagenesis qualifies as having a sufficient safety record this is not so for new genome editing techniques. They are therefore not covered by the mutagenesis exemption [ECJ Case 528/16 paras 46–53]. This means that in the EU all new synthetic biology techniques involving transgenesis and non-physical and non-chemical mutagenesis are within the scope of the regulatory oversight. The EU legislator has the possibility to modify the exemptions and decide what applications of synthetic biology are safe enough to be listed as exempted techniques, or subject certain techniques to less stringent tools of oversight, such as prior notification or ex post monitoring and reporting instead of prior authorisation.

In the United States, certain synthetic biology products may not be covered by existing product-related legislation. The US Plant Protection Act, for instance, only covers plants if a plant pest, such as an agrobacterium, was used to introduce the genetic material. This would not cover new synthetic biology techniques which use CRISPR-Cas9 or other pathways to insert a gene or otherwise modify the organism (Bergeson *et al.*, 2015, 45). The US Department of Agriculture issued a statement in March 2018 that it would not regulate plants developed through genomic editing techniques which are indistinguishable from plants that could be developed through traditional breeding techniques ([https://www.aphis.usda.gov/aphis/ourfocus/biotechnology/brs-news-and-information/2018_brs_news/pbi-details][https://www.usda.gov/media/press-releases/2018/03/28/secretary-perdue-issues-usda-statement-plant-breeding-innovation]).

Another example is the dengue transmitting mosquito *Aedes aegypti* into which a lethal gene was inserted that through reproduction could lead to a reduction in populations. The modified mosquito was initially determined to be covered by the US Federal Food, Drug and Cosmetic Act (FFDCA) as an animal drug, analogous to other drugs used for animal population control. However, unlike other such drugs, the mosquito was intended to be released in the wild and used for the purpose of addressing human disease, raising questions about the appropriateness of FDA jurisdiction (Bergeson *et al.*, 2015, 20). In 2017, the FDA announced that products "intended to function as pesticides by preventing, destroying, repelling, or mitigating mosquitoes for population control purposes" would not be considered "drugs" under the FFDCA, but would instead be regulated as "pesticides" by the EPA (FDA, 2017a). In 2018, the EPA opened public comment on an application for an experimental use permit for genetically engineered *Aedes aegypti* mosquitos ([https://www.epa.gov/pesticides/epa-reopens-public-comment-period-application-experimental-use-permit-combat-mosquitoes]).

In Japan, the Advisory Panel of GMOs of the Minister for the Environment has proposed that any product not categorised as genetically engineered under the Cartagena Protocol shall be exempted from the existing regulation. The Panel suggested that this would include any product created through genome editing that does not involve introduction of foreign nucleotides, such as deletions, as well as any product developed through introduction of material from species which could naturally cross with the host organism. The proposal is open for public comment and has not been formally adopted (USDA, 2018a).

In New Zealand in 2014, the Environmental Protection Authority decided that plants produced via gene editing methods, where no foreign DNA remained in the edited plant, would not be covered by GMO legislation. However, this decision was successfully appealed to the High Court, which overturned the decision on the basis that creating exceptions to the GMO regulations was a political decision and not within the power of the Authority [Sustainability Council v EPA, 2014, 69] (Kershen, 2015). In reaching its decision, the Court affirmed the applicability of the precautionary approach based on the scientific uncertainty related to environmental effects of rapid changes caused by the technology [Sustainability Council v EPA, 2014, 68]. Following this decision, all products of gene editing are currently captured within the scope of legislation in New Zealand (Fritsche et al., 2018).

A number of additional countries are currently considering what applications of genetic modification fall within the scope of risk assessment frameworks for GMOs. Chile, Brazil, Israel, Argentina and Australia, among others, have adopted or introduced regulations clarifying whether products of genome editing can be considered GMOs for the purpose of risk assessment regulation (Duensing *et al.*, 2018). In general, the likelihood of biotechnology products falling within the scope of existing regulation relates to the use of recombinant DNA and the degree of change to the host DNA sequence.

2.3.2 Risk/benefit assessment of novel organisms

Synthetic biology applications challenge existing risk assessment paradigms due to their potential to express novel traits, persist in the environment, and cross geographic and political boundaries (NASEM, 2016a). Existing risk assessment paradigms for genetically engineered organisms have largely been developed and used to assess the risks from two novel traits in plants: insect resistance and herbicide tolerance. Novel synthetic biology and gene drive applications will have traits that differ quite drastically from these. While the overarching risk assessment process may not change, specific steps within risk assessment will need to be tailored to these new applications. Decisions will have to be made concerning how to change risk assessment approaches to adequately assess the potential harm caused by organisms that have not previously existed (NASEM, 2016a; Hayes et al., 2018). New concerns may arise, for example relating to the uncertainty and difficulty of conducting a complete environmental risk assessment without environmental release. Furthermore, the values-laden judgments inherent to the risk assessment process (Section 3.4.3) will receive extra scrutiny, given the

novel and controversial nature of synthetic biology (Stirling, Hayes & Delborne, 2018; Thompson, 2018).

A major characteristic of risk assessment for traditional GMOs is the familiarity or comparison approach. This has been described as a "comparison of the characteristics of the GMO(s) with those of the non-modified organism under corresponding conditions of the release or use" and is intended to help identify "the particular potential adverse effects arising from the genetic modification" [Directive 2001/18/EC Annex II sec. B. 1st indent and sec. C. 2.1]. There are suggestions that the comparison with parental and/or non-modified organisms loses validity where synthetic biology does not only marginally modify an organism but can create essentially new ones (Winter, 2016b). A proposed alternative to the comparison approach is use of a set of tests following a step-by-step and case-by-case approach to information generation before the release of the modified or new organism is approved (see above Section 2.2.1.3).

Applications of synthetic biology can create irreversible effects. In some cases, as in use of engineered gene drives to eradicate a species from a certain habitat, irreversibility could be seen as part of the intent. There have been calls for development of effective reversal drives as part of regulatory requirements for engineered gene drives (Oye et al., 2014). Such risk management measures could provide a means to address indirect or unintended environmental impacts, but even if effective, they would not address intended impacts. Moreover, permanent damage could be caused before the reversal drive reached all members of the population (Esvelt et al., 2014).

2.3.3 Transboundary movement

International and national law have established mechanisms for managing transboundary movement of genetically modified organisms and potentially hazardous substances as well as principles for addressing transboundary harm (see Section 2.1.2). To some extent these existing structures provide a framework applicable to transboundary impacts of synthetic biology. However, certain applications of synthetic biology, including engineered gene drive systems, create questions related to coverage and implementation of these frameworks.

Two types of transboundary movements can be envisaged when considering synthetic biology: unintended and intended. Some applications of synthetic biology focus on particular geographies, contained within country borders. This is the case for applications against invasive species that intend to suppress those species locally but are not intended to have such effect on a global scale. If those applications were to be moved across borders, it would be an unintended or illegal transboundary movement [for definitions of unintended or illegal transboundary movement, refer to Decision VIII/16 of the Cartagena Protocol on Biosafety and its annex Operational definitions of the terms "unintentional transboundary movement" and "illegal transboundary movement"]. This could happen through natural dispersal of modified individuals, or through human transport (intentional or unintentional). For unintended transboundary movement, there are existing governance frameworks. Under Article 17, the Cartagena Protocol requires countries to notify other countries that might be affected by an unintentional transboundary movement that may have an adverse effect on biodiversity.

Another set of the technologies, approaches and tools are intended to move across boundaries. For example, the vector control applications of engineered gene drive for malaria (see Chapter 6) are intended to address vector movement across different countries, as this would be an important factor for success. Several recent reports looking at engineered gene drive for malaria control have raised the importance of regional approaches (James et al., 2018), or coordination and communication between neighbouring countries (NASEM, 2016a). The Cartagena Protocol requires states from whose territory organisms are intentionally moved across borders to obtain advance informed agreement from the importing state. However, this provision was developed in the context of transboundary import and export, and it is not clear how it applies to intended or anticipated spread of modifications across borders (NASEM, 2016a).

Transboundary damage can create particular problems for compensation or restitution. The Supplementary Protocol applies to damage resulting from both intentional and unintentional transboundary movement as well as illegal transboundary movement, and requires Parties to mandate response measures in the event of damage [Arts 3, 5]. However, the application of civil liability in the event of transboundary damage is largely left to be determined under domestic law. This can raise questions relating to proving causality and quantifying harm, particularly where the modified organism does not cause direct economic or environmental damage.

These issues are in some ways analogous to the governance of biological control agents. In that context, they have been addressed through discussion and harmonisation of measures at the regional level (Bateman, Sulaiman & Ginting, 2014). The African Union has started looking at regional harmonisation around the possible use of engineered gene drive for malaria control (NEPAD, 2018).

In addition to the regulatory question, the potential of intended or unintended transboundary movement raises challenges for stakeholder engagement, to ensure that public consultation can be carried out at the appropriate level.

2.3.4 Digital sequence information

The growing use of genetic information derived from digital sequencing in synthetic biology creates uncertainty for access and benefit-sharing regimes (see Section 6.6.1 for a description of technological advances in digital sequence information). There have been numerous studies examining the impact digital sequence information and synthetic biology may have on access and benefit-sharing agreements around genetic material (Bagley & Rai, 2014; Bagley, 2016; Welch et al., 2017; Wynberg & Laird, 2018b; *see also* Table 1.1).

At the CBD, where "genetic resources" were primarily envisioned and defined as genetic material, a process is now underway to respond to the potential implications of the use of digital sequence information on CBD objectives [CBD COP13 Decision 16; COP14/L.36].

An *ad hoc* technical expert group on digital sequence information on genetic resources was established to consider the potential implications of the use of digital sequence information on genetic resources for the CBD.

Submissions from countries and other stakeholders to the CBD expert group show the range of perspectives on considering digital sequence information "genetic resources." For certain non-governmental organisations, such as the Third World Network, not regulating such information under the CBD could "economically and culturally undermine indigenous peoples and local communities, thereby negatively impacting the conservation and sustainable use of biodiversity." They point to the use of synthetic biology to produce vanilla and vetiver as examples of the potentially disruptive impact on farmers and other local actors (AHTEG, 2018b). For research organisations such as the UK Natural History Museum, Royal Botanic Gardens Kew, and Royal Botanic Gardens Edinburgh, there are potentially negative implications in regulating access to digital sequence information. They highlight the value of digital sequence data in the public domain for biodiversity conservation and sustainable management and the impracticability of asking open-access international databases to regulate the use of digital sequence data. It is also put forth that the current mechanism for sharing digital sequence information might already be considered the equivalent of a global multilateral benefit-sharing mechanism (AHTEG, 2018a). Some researchers have argued that including digital sequence information under the Nagoya Protocol would create a global damper on research(Kupferschmidt, 2018).

A scoping study commissioned by the CBD found that the use of information on genetic resources, including in synthetic biology, could create opportunities for new forms of non-monetary and monetary benefit-sharing (Wynberg & Laird, 2018). At the same time, the study noted the risk that access to digital sequence information would allow researchers to look at the genetic or biochemical composition of genetic resources without having to physically access the resources themselves, which could undermine existing approaches to access and benefit sharing.

If the genetic information is deemed to fall within the scope of "genetic resources" in the CBD, the challenge will be defining whether and how the principle of sovereignty over genetic resources and the system of access and benefit-sharing based on this principle can address these vastly different dynamics. In his book on genetic resources as natural information, Ruiz (2015) notes that: "Inasmuch as information constituents can be stripped from their physical medium in biological samples, attempting to institutionalize controls over the flow of information, disembodied at different moments, by different actors, and in different places, is not only impossible but absurd." Ruiz advocates a conceptual framework for ABS based on the economics of information, as well as an alternative mechanism for ABS that is multilateral, non-contractual and focused on fairness and equity in the sharing of monetary benefits. Such a multilateral benefit-sharing mechanism is possible under Article 10 of the Nagoya Protocol. In discussions under this article, at least one country – Argentina – has noted that a global multilateral mechanism may be useful for the use of digital sequence information (SBI, 2018).

The evolving technological, legal and institutional context surrounding the exchange and use of digital sequence information (DSI) for synthetic biology and genomic research may affect access to ABS frameworks under the ITPGRFA (Welch et al., 2017). The availability of sequence data through decentralised data libraries and organisations may challenge the multilateral system set up by the ITPGRFA (Welch et al., 2017). Other factors including partial sequence combinations, and the fact that the same sequence may occur in multiple organisms create further questions for ABS (Welch et al., 2017).

2.3.5 "Do-it-yourself" (DIY) biology

The tools associated with synthetic biology are becoming increasingly accessible to private actors, including actors who may not have the backing of an established institution. This raises governance questions as well as some public concern (Charo & Greely, 2015). Many of these concerns may be based on an inaccurate understanding of the activities and capabilities of community laboratories (Kuiken, 2016). However, as with any decentralised activity, the DIY aspects of synthetic biology research create certain challenges to traditional models of governance.

One concern centres on safety. DIY biologists may not be held to the same standards of safety as formally trained biologists (Garrett, 2013). In some jurisdictions, licensing requirements on laboratory biologists, including training in safety and ethics, may not apply to community laboratories (Kolodziejczyk, 2017). However, in Germany and other countries in Europe, community laboratories, like other laboratories, need licenses to undertake experiments involving genetic engineering (Seyfried, Pei & Schmidt, 2014). In all countries, biosafety regulations and risk assessment and management procedures covering synthetic biology activities – including requirements relating to notification, authorisation, containment, transfer and monitoring – would apply to DIY biologists as well as formal labs. The DIY biology community has also developed its own safety standards (Guan et al., 2013) as discussed above, and continues to evaluate their effectiveness and develop additional resources associated with biosafety and biosecurity (Yassif, 2017).

Where they are held to the same or similar licensing standards as formal laboratories, community laboratories will also be required to obtain insurance. In some countries, such as Tanzania, all operators engaging in activities involving genetic modification are required to carry insurance [Tanzania Biosafety Regulations, 2009, § 35(1)]. In other countries, DIY biologists operating outside an institutional setting may not have explicit insurance requirements, though many of the labs may carry this type of insurance regardless. This creates a potential problem if something does go wrong, as community biologists may not have the resources to cover costs of compensation or remediation.

As DIY biology becomes more accessible to users not associated with a particular institution, this may raise challenges for enforcement of biosafety and environmental regulations against actors with bad intent. While the community's own regulations

may support safe practices among well intentioned operators, informal or illegal operators with bad intent may be difficult to identify and hold liable (Garrett, 2013). However, there are still limits on the capability of community laboratories to create organisms that would cause significant environmental damage, and to date there has been no evidence of attempts or intent to do so (Lentzos, 2016).

Much of the concern around DIYbio centres relates to questions of biosecurity. These questions are outside the scope of this assessment, though there has been some thinking in the biosecurity context that could be relevant to governance of DIYbio to prevent environmental impacts. Kelle (2009) proposes a "5P" strategy that outlines five points of intervention for managing risks: principal investigator, project, premises, provider (of genetic material) and purchaser. At each of these points, measures ranging from awareness raising and education to industry codes of conduct to national and international laws and regulation could be used to prevent misuse (Kelle, 2009).

An issue hardly discussed is the application of ABS regulations to DIY biology. Any rules user states may have established to ensure compliance with pertinent provider state regulation also apply to DIY synthetic biologists. But DIY biologists may not be aware of this, and it could be difficult for user state authorities to supervise their research and development in termsof ABS.

2.3.6 Research and governance capacity

Emerging economies represent significant potential markets and research centres for synthetic biology as well as providers of genetic material that may be used. However, capacity varies across jurisdictions, with implications for both research and governance.

In emerging economies, research capacities across disciplines and departments with regards to synthetic biology are underdeveloped. Developing and upgrading research and development facilities represents significant capital investment. There is consensus that emerging economies require support in this regard [Cartagena Protocol art. 22] but the form and nature of capacity needed is still unclear. Advanced applications require advanced skills and capital which can delay synthetic biology development and the deployment process. The African Union recognises the need for strengthening the capacity on the continent in order to harness the potential benefits of these developments while being able to ensure that those are co-developed with African scientists (African Union, 2018). Recent growth in digital innovation in Africa and Asia indicate potential for technological entrepreneurship. In 2018, teams from Uganda, Egypt, Singapore and Pakistan, among others, participated in the International Genetically Engineered Machine Championship (iGEM.org).

Emerging economies also represent potential markets for synthetic biology applications and products. Certain types of technology may be nationally or regionally prioritised based on context and needs (African Union, 2018). In Africa for instance, production of synthetic biofuels may have immediate environmental, social and economic benefits (Stafford et al., 2018).
There is evidence of gaps in legal frameworks and capacity for regulatory oversight in many developing countries. Few countries have enacted biosafety laws that could act as reference points for synthetic biology development and diffusion (Figure 2.1). Of significance is the lack of or inadequate provisions for post-release phases. Governments also faced the challenge of balancing a precautionary approach with the potential economic benefits of synthetic biology applications (Kingiri & Hall, 2012).

Reduced technical and regulatory capacity made worse by porous national and regional borders raise questions of biosafety and potential misuse of synthetic biology. There have been calls for harmonisation of biosafety- and trade-related policies with clear guidelines for deployment of synthetic biology applications and products at respective national levels to enhance responsible and productive synthetic biology piloting, products release, monitoring and surveillance (Escaler et al., 2012).

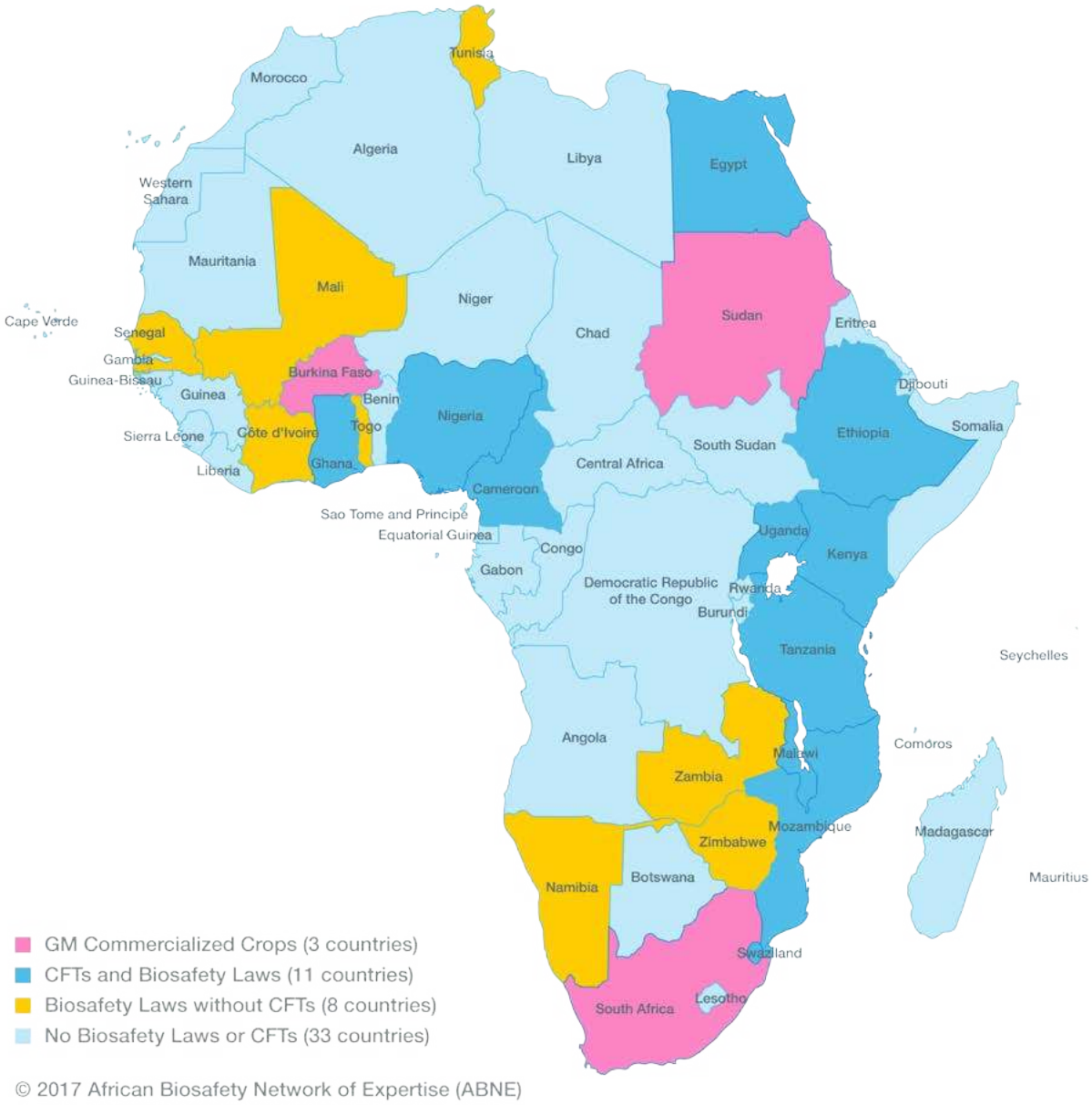


Figure 2.6 Biosafety Laws in Africa. Adapted from a graphic by the African Biosafety Network of Expertise.

2.3.7 Funding and financial flows

The funding sources and financial flows associated with synthetic biology (Section 1.6) have influenced the discourse around governance. Availability and access to funding drives innovation. While some private organisations, such as the Gates Foundation-funded Target Malaria project, fund work pursuing the safe and effective use of engineered gene drive systems, most funding comes from public sources.

In Europe, funding for synthetic biology has primarily come from public funding organisations such as the Swiss National Science Foundation, UK Research Councils and the Netherlands Organisation for Scientific Research (Pei, Gaisser & Schmidt, 2012). In the United States, there are few publicly funded research programmes outside military programmes, such as the US DARPA Safe Genes Program (DARPA, 2018d). Before 2008, the US federal government invested relatively small amounts in synthetic biology. By 2014, it had invested approximately US$ 819 million in synthetic biology research (WWC, 2015). Since 2012, the majority of that funding came from Department of Defense initiatives (see Chapter 1). A recent exception is approximately US$ 2 million

from the US Department of Agriculture's National Institute of Food and Agriculture (NIFA) for research on the implications of gene edited technologies, including one project explicitly focused on engineered gene drive systems in agriculture (USDA, 2018b).

There have been calls for increased funding for research into ethical, legal and social issues relating to synthetic biology. A 2012 review of European public funding organisations showed that where such funding is available, there can be problems in linking funding opportunities with the research community (Pei et al., 2012).

Concerns have been raised about synthetic biology funding patterns, partly regarding the agenda behind the funding, and the purpose, or alternate purposes, to which the technology and its applications might be used (Lentzos, 2015; Kuiken, 2017; Reeves et al., 2018). Concerns range from the power funders have to determine the trajectory of research to problems of conflict of interest in scientific research, whereby the objectivity of researchers is compromised – or perceived to be compromised – by sources of funding or other institutional commitments (Krimsky, 2004, 2013). In addition, synthetic biology's technical and institutional connections to agricultural biotechnology create discursive links to critiques of the political economy of first-generation genetically-modified organisms (Charles, 2001; Schurman, Kelso and Kelso, 2003; Worthy et al., 2005; Kleinman & Vallas, 2006; Delborne, 2008; Kinchy, 2012). As such, concerns have been raised that synthetic biology will benefit private over public interests, continue enclosures of genetic commons through aggressive intellectual property practices, concentrate power in the hands of elites, and undermine more holistic and traditional approaches to sustainability (e.g. ETC Group, 2018). More research is required to understand where and under what conditions these concerns may actualise, and how to prevent them from doing so (Pottage, 2006; Calvert, 2008; Lawson & Adhikari, 2018).

2.3.8 Moral hazard

Synthetic biology creates a fundamental challenge for risk assessment and conservation governance more broadly in the form of what is called moral hazard. "Moral hazard" means that new technologies may correct the symptoms of, and provide an excuse not to address, more fundamental socio-political failures which caused the symptoms in the first place. For example, climate change caused by increased emissions of greenhouse gases into the atmosphere is projected to cause changing weather patterns including increased droughts potentially affecting food production. Fundamental change would require that emissions are drastically reduced. Moral hazard occurs if new technologies, such as drought-resistant crops, create excuses for decision makers not to implement mitigation policies to prevent droughts. In this example, even if synthetic biology can lessen the severity of certain consequences from climate change-induced droughts, the vast number of consequences caused by such droughts simply cannot be addressed through synthetic biology alone – the fundamental problem needs to be addressed. The same applies to engineered gene drive technology. If applied as a means of nature conservation it may foster a vision that traditional habitat and species protection can be replaced by just making species and habitats resilient to new stresses.

2.3.9 Engaging with multiple perspectives and ethics

As has been highlighted in Chapter 1, there are a number of ethical questions raised by synthetic biology. Ethics are value systems that shape the perception, assessment and management of a technology. Ethics also shape governance systems in multiple ways. Many governance systems are based on norms and concepts deriving from ethics. Ethical considerations are behind calls for limits on certain applications of synthetic biology, such as use of gene editing on human beings, which can influence national and international law (e.g. Convention on Human Rights and Biomedicine of the Council of Europe; Grubb, 1994). Ethical considerations will influence the scrutiny of risk assessment, the determination of acceptable risk, and the weighing of benefits and risks in decision making related to synthetic biology research and introduction into the environment.

There is wide recognition that ethical arguments are important to take into consideration when considering

synthetic biology applications and they need to be clearly framed when responding to the concerns of different cultural traditions and political orientations within and between particular communities or regions (Winter, 2016a; Zetterberg & Edvardsson Björnberg, 2017). The ethical debate about science and technology is often done in absolute terms at a given time, but increased experience and exposure can change perspectives, sometimes in favour of technology and sometimes against it (UNESCO, 2015). The diversity of moral perspectives and values inform decision making, but also creates a challenge for regulation.

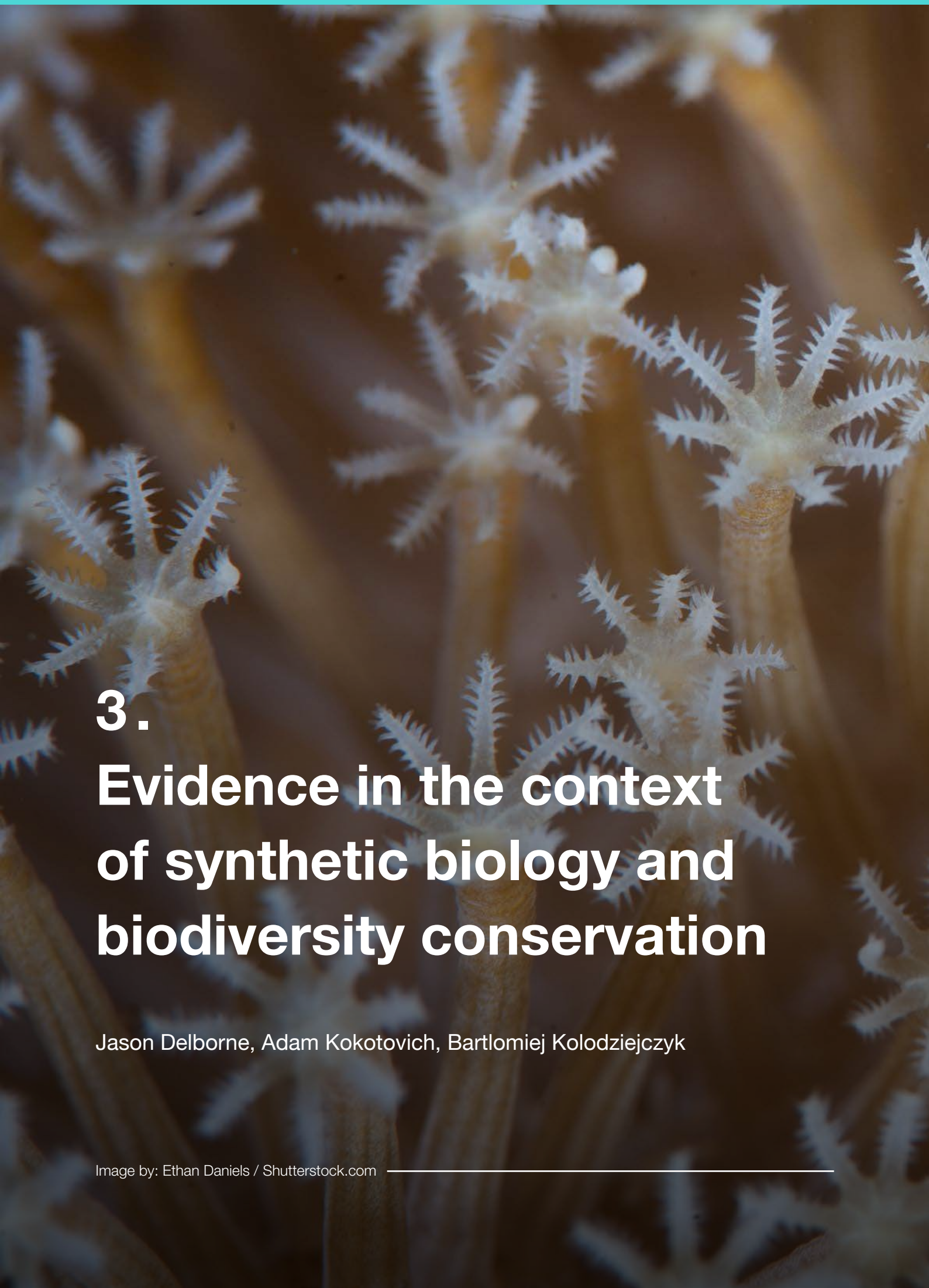
Scientists themselves have questioned their practice in response to ethics with normative instruments such as the UNESCO World Conference on Science Declaration on Science and the Use of Scientific Knowledge [1999] that calls for responsible science and its interaction with society's values. Synthetic biology researchers are becoming increasingly cognisant of the ethics and value-based discussion about synthetic biology and how its potential application as well as the research itself can question values in society. This recognition has been translated to action with the integration of "ethical, legal and social implications" (ELSI) into research networks and programmes (Synbiosafe, 2018) and a growing interaction between ELSI experts and synthetic biology researchers (DARPA, 2018d).

Even in the absence of guidelines or regulatory requirements, researchers and scientific associations drawing on field experience and literature argue that transparency and openness are the foundation for ethical engagement (Esvelt et al., 2014; NASEM, 2016a; Resnik, 2018). They agree that engagement should ensure that evidence and uncertainties about both potential risks and benefits are shared with the public.

Engagement also needs to be responsive to input and information received from stakeholders. The Royal Society dialogue on gene editing (Van Mil, Hopkins & Kinsella, 2017) showed the importance for stakeholders of ensuring the engagement was not a box-ticking exercise and was going to be taken into consideration by policy makers (Van Mil, Hopkins & Kinsella, 2017). Organisations such as LEAP Synthetic Biology made calls to use deliberative dialogues to ensure that communities' perspectives would be taken into consideration seriously during policy-making processes (Ritterson, 2012).

While the dialogue might enable discussion of different values, perspectives and understanding of evidences, researchers recognise that it is important to build mutual understanding in order to achieve a meaningful dialogue (UNESCO, 2015). Practitioners also recognise the need for a structured and continuous engagement and the establishment of clearer engagement pathways (NASEM, 2016a).

Although researchers' commitment to engagement is critical, it is not sufficient. There is also a need for national governance mechanisms to provide guidelines about the remits and scope of the engagement and of stakeholders' participation in decision making so that engagement can be aligned (NASEM, 2016a). While there are existing guidelines for public consultation (EFSA, 2018), there have been criticisms from concerned NGOs and scholars about bias in engagement, particularly where it is undertaken by the proponent of the technology, as well as limited identification of who is entitled to give consent and how consent is sought (Unknown, 2014; Bäckstrand et al., 2010).



3.
Evidence in the context of synthetic biology and biodiversity conservation

Jason Delborne, Adam Kokotovich, Bartlomiej Kolodziejczyk

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3.1 What does it mean to be "evidence-based"?

This assessment is charged with the task of conducting an evidence-based examination of the potential benefits and risks of synthetic biology and engineered gene drive applications to biodiversity conservation. In the context of a contentious and emerging field such as synthetic biology, the idea of "evidence-based" requires examination. This chapter discusses evidence with the goal of fostering productive conversations on the science and governance of synthetic biology and sets the stage for this assessment. The chapter has three sections that follow this introductory discussion. Sections 3.2 and 3.3 introduce the topic of scientific evidence by discussing peer review, reproducibility, replicability and uncertainty. Section 3.4 explores the broader factors that influence the creation and synthesis of scientific evidence. As a whole, the chapter offers a nuanced view of the challenges and importance of integrating scientific knowledge into decision making.

What explicitly is meant by an evidence-based assessment? "Evidence-based" is an often-cited goal for assessments and decision processes surrounding technology and the environment, but what this exactly means can vary. On the one hand, it can be used to emphasise that decisions on the use of technologies need to be informed by empirical studies examining their efficacy, potential benefits and risks. On the other hand, the term evidence-based can be understood as an attempt to remove values and politics from assessments or decision-making processes. This latter interpretation, rarely feasible, fails to acknowledge the subjective judgements and values that inform assessments and decision making. This could in turn privilege the values of scientific experts, which may differ from those of other stakeholders and publics (Sarewitz, 2015). Scholars of science and technology policy have argued that it is far more responsible and productive to acknowledge the role of values within such processes and to use an appropriate form of deliberative engagement to sort through evidence, uncertainty and preferences (Jasanoff, 2003; Rowe & Frewer, 2005; Pielke Jr, 2007). This assessment thus adopts the first interpretation: decisions on the use of technologies need to be informed by empirical studies examining their efficacy, potential benefits and risks. The chapter therefore explores the factors that impact the production and synthesis of scientific evidence and how they were navigated within this assessment, including how deliberation was used throughout the process. While the focus of this chapter is on the factors impacting scientific evidence, other types of evidence and concerns – as discussed in the previous chapter on governance – also need to be incorporated into decision making surrounding synthetic biology and gene drive.

To inform the discussion of how evidence, values and deliberation shape this assessment, the scholarly field of responsible research and innovation provides four principles: anticipation, inclusion, reflexivity and responsiveness (Stilgoe, Owen & Macnaghten, 2013). These principles were originally developed to inform the governance of emerging technologies, and their significance for an assessment like this is apparent.

Anticipation refers to the need to predict the potential futures of synthetic biology, including engineered gene drive systems, so as to guide them towards desired ends and away from undesired ones – epitomised by the Resolution that mandated this assessment.

Inclusion highlights the need to pay close attention to who is involved in decision-making processes, including, in this assessment, the process of deciding its scope and synthesising relevant evidence. The decision to conduct the assessment was an inclusive one, since it was IUCN's diverse and representative membership of 1,303 government, civil society and indigenous peoples' organisations that passed Resolution WCC-2016-Res-086. For the assessment itself, as referenced in the accompanying statement of principles (Front Matter) and the process flow (Figure 1.10), inclusion has been achieved through: (i) disciplinary, gender and geographical diversity being considered in selecting the group to complete the assessment, which, unusually, falls under the mandate of all six IUCN Commissions as well as the Director General, and (ii) opening this assessment to external peer review from all IUCN Members and anyone else who wished to participate.

Reflexivity denotes the need to be aware of the key assumptions and judgements being made within this

assessment. This includes, for example, reflecting upon the assumptions informing how evidence is selected and synthesised – a process that motivated the inclusion of this chapter. Deliberative processes were used during the creation of this assessment to foster this reflection, and peer review processes (Section 3.2.1) offered further opportunities for reflection inspired by external perspectives.

Responsiveness embraces the need to be open to changing in response to insights garnered through inclusive deliberation – embodied in the assessment's process for responding to peer review, including the transparent documentation of responses to all comments (see iucn.org/synbio).

These pillars of responsible research and innovation serve as a reminder that seeking "evidenced-based" decision making is about more than the quality of the data and the identification of key experts; it requires careful attention to the processes through which evidence is generated, gathered and considered in decision processes that must reflect the complexity of society itself. Indeed, as Ascher, Steelman and Healy (2010) argue, knowledge for environmental decision making extends beyond formal scientific knowledge to local and indigenous knowledge, as well as knowledge about public preferences.

3.2 What is scientific evidence?

Scientific evidence derives from a rigorous process that serves to either support or counter a theory or hypothesis (Popper, 2005). The significance of scientific evidence often relies on collection and analysis protocols (Bilotta, Milner & Boyd, 2014) and is based on the results of quantitative (e.g. statistical) and qualitative (e.g. textual) analysis. Scientific evidence is generally expected to be empirical; however, standards may vary depending on the field of inquiry (Becker, Bryman & Ferguson, 2012). Because of this variation, some scholarship has shown how standards of scientific evidence are defined and negotiated among the participants in different disciplines (Knorr-Cetina, 1999), suggesting that there is no absolute or incontestable standard for evidence in any field (Collins, 1983), especially an emerging field such as synthetic biology. Nevertheless, the existence of scholarly traditions in ecology, molecular biology, science and technology studies, and ethics provide guidance in evaluating the power and importance of scientific evidence in emerging fields (Knorr-Cetina, 1999; Sismondo, 2010).

Scientific evidence drawn from a single study will rarely provide a meaningful answer to a given question. As such, it is important to examine and weigh the pieces of scientific evidence from a broader body of research to make an informed conclusion (EFSA Scientific Committee, 2017). To be considered established, scientific evidence must be consistent and generally accepted by the broader scientific community, though traditions of scepticism and dissent are also important in visions of advancing scientific understandings (Kuhn, 1970; Merton, 1973; Delborne, 2008).

Scientific communities often distinguish between a hypothesis and a theory. Generally, a hypothesis is a proposed explanation, often based on prior knowledge or basic experiments, that can be tested through further experiments and observations. Further data are required in order to confirm or reject a hypothesis, whereas a theory is a widely accepted concept supported by a substantial body of evidence (Sutton & Staw, 1995). While some synthetic biology theories are already supported by significant bodies of evidence, many hypotheses are yet to be proven due to the relative immaturity of the field. The scientific community has mechanisms for overturning accepted theories based on new evidence. This is often a complex process – not solely determined by the quality of evidence – that is influenced by disciplinary norms, challenges launched from related disciplines, changes in cultural understandings, and other social factors (Kuhn, 1970; Longino, 1990; Gieryn, 1999).

In certain cases, it might not be possible to draw conclusions based on observations because the work in question has not been done; in such cases, mathematical models may be used to support informed decisions (Knight et al., 2016). Using mathematical models and simulations is of specific relevance to synthetic biology and biodiversity conservation, where experimental evidence is limited. For instance,

computational models have recently been used to model the potential spread and persistence of engineered gene drive organisms without actually releasing them into the environment (Unckless, Clark & Messer, 2016; Eckhoff et al., 2017; Noble et al., 2018). Such efforts further scientific understanding without producing any direct environmental impact. Of course, models never perfectly capture ecological, biological and social contexts, meaning that experimental evidence will be required to challenge model predictions and solidify scientific understanding. Negotiating such experiments is complex and may be contested in the field of synthetic biology, where the testing of new technologies such as gene drive organisms creates the potential for environmental impact beyond the boundaries of a field trial (NASEM, 2016a). Making decisions about such field trials thus triggers governance processes that weigh the needs for empirical data against the potential for unintended ecological adverse effects.

3.2.1 Peer review

Peer review is a process meant to ensure research quality, validity and appropriate presentation of scientific evidence (Gannon, 2001). Peer review serves as a key gatekeeping mechanism to expose proposed scientific evidence to critique by experts (Merton, 1973). The peer review process is carried out by employing experts with relevant expertise (Voight and Hoogenboom, 2012) and is designed to ensure the appropriateness of data collection and analysis and to prevent scientific fraud (Kelly, Sadeghieh & Adeli, 2014).

The peer review process is not perfect (Mulligan, Hall & Raphael, 2013). Most importantly, the process relies on existing knowledge and tends to assess the validity of work based on previous studies or what is generally accepted by the scientific community (Gannon, 2001). Findings that challenge existing understanding thus might be criticised and rejected as poor or incorrect (Kuhn, 1970). Peer review can also be challenging when credible studies offer conflicting interpretations or conclusions or when multidisciplinary work makes it challenging to find reviewers with an adequate range of expertise (Langfeldt, 2006).

This assessment included a peer review process for solicited independent experts, IUCN stakeholders and interested members of the public to provide feedback – including comments, critiques and suggestions for consideration of additional evidence. In this review process neither authors' nor reviewers' identities were hidden, making this a so-called "open" peer review. The application of open review was guided mainly by the technical group's principles of transparency, inclusivity and consultation, as well as IUCN's generally accepted practices. The potential disadvantage of open review is that some reviewers may limit the feedback provided due to concerns of being identified personally with their comments. The wide range of supportive and critical comments received on the draft of this assessment, however, offers some reassurance that this disadvantage of open review was minimal. This final draft of the assessment thus represents the technical group's best effort to incorporate and respond to the comments received (to view all comments and responses, see iucn.org/synbio).

3.2.2 Norms of reproducibility and replicability

Reproducibility and replicability are two concepts that play significant roles in evaluating the quality and reliability of scientific work (Stodden, 2009; Jasny et al., 2011). Scientific evidence is valued more if the same observations and conclusions can be drawn from multiple independent studies. In emerging disciplines, such as synthetic biology, the lack of a sufficient number of independent studies can be challenging.

In the biological sciences, independently reproducing a set of observations can be costly and time consuming. For instance, reproducing gene drive observations in wild populations would take generations. It might be feasible for species with short lifespans, but becomes more challenging (given the pressure for rapid scientific results) for longer-lived species. Reproducibility, while highly valued, has generated significant recent controversy in a number of fields where researchers have found it difficult or impossible to reproduce accepted findings (Arrowsmith, 2011; Begley & Ellis, 2012; Baker, 2016).

3.3 Engaging with uncertainty

As with most emerging technologies, uncertainty complicates efforts to assess the impacts of synthetic biology. Uncertainty concerning the impacts – intended and unintended – of synthetic biology applications may be caused by a variety of factors, such as the limitations of modelling or low levels of empirical evidence. In considering the release of gene drive altered organisms into the environment and their impact on wild populations, for example, uncertainties are relevant to questions involving the effectiveness of the engineered gene drive, its stability over time, the fitness costs of the genetic constructs, and effects on non-target organisms (NASEM, 2016a). Scholars have developed many different methodologies to evaluate and classify uncertainty (Milliken, 1987; Warmink et al., 2010; Hayes, 2011). Here we provide a typology of sources of uncertainty and provide a four-quadrant model to help classify the degree of confidence in evidence.

Hayes (2011) identifies four sources of uncertainty: epistemic uncertainty, variability, linguistic uncertainty and decision uncertainty. *Epistemic uncertainty*, likely the most common in discussions about emerging technologies, is the uncertainty associated with the production of knowledge. It is the result of imperfect knowledge regarding something that is in principle knowable, and, therefore, additional research can reduce this type of uncertainty. *Variability* refers to the unavoidable uncertainty caused by natural variation or inherent randomness. Unlike epistemic uncertainty that can be decreased with further study, this type of uncertainty cannot be reduced. Variability is relevant for this assessment because biodiversity conservation never happens in a perfectly uniform environment. *Linguistic uncertainty* results from the imprecision of language and has five causes: vagueness, context dependence, ambiguity, indeterminacy and underspecificity. This assessment seeks to reduce linguistic uncertainty by defining key terms in the Glossary and presenting a conceptual framework for evidence-based decision making in this chapter. *Decision uncertainty* occurs when there is ambiguity concerning how to quantify or compare social objectives to inform a decision (e.g. how to weigh relative risks and potential benefits in the decision to permit a field trial of a gene drive modified organism). The Governance chapter of this assessment addresses the decision uncertainty surrounding the application of synthetic biology and gene drive to biodiversity and conservation.

This assessment engages an explicit framework to describe the certainty of key messages. A four-quadrant diagram qualitatively describes the degree of certainty associated with a finding or idea (see Figure 3.1), drawing upon the Intergovernmental Panel on Climate Change (IPCC) uncertainty classification (Moss & Schneider, 2000) and the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services (IPBES) classification (IPBES, 2016b). In this framework the degree of certainty is dependent upon the quantity and quality of evidence, in addition to the level of agreement on that evidence. *Well established* refers to a finding that is supported by a meta-analysis or multiple independent lines of evidence. *Established but incomplete* refers to a finding that is generally agreed upon but supported by a limited number of studies. *Competing explanations* denotes a situation where many independent studies exist but different conclusions are drawn from them. *Speculative* findings are where there is low consensus on the limited evidence that exists – this quadrant represents areas with major knowledge gaps.

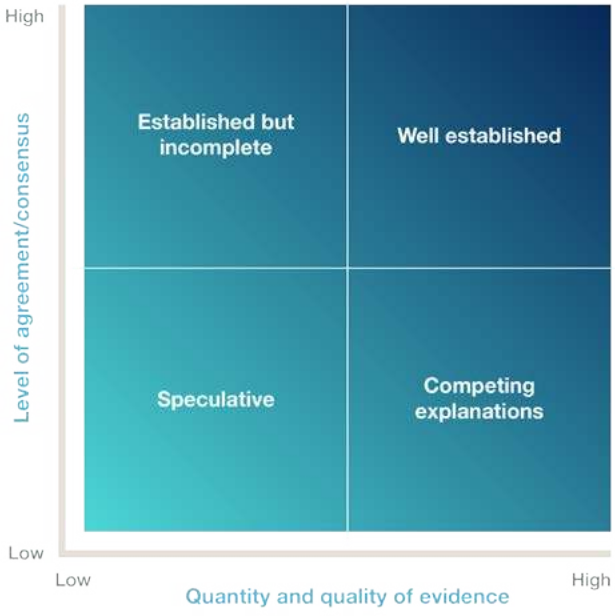

Figure 3.1 Qualitative uncertainty terms. Synthesis of Moss and Schneider (2000) and IPBES (2016).

In contrast to the epistemic uncertainty suggested by Figure 3.1, so-called "unknown unknowns" are the most challenging type of uncertainties due to their unknowable nature, which makes them difficult to address in a risk assessment or even a precautionary approach (see Section 2.1.1). This uncertainty can result from inherent randomness and a lack of evidence to even conceive of and characterise what is known and not known. As such, effective strategies to anticipate unknown unknowns do not exist. This type of uncertainty has to be dealt with as evidence emerges. While specific cause and effect relationships are generally not established with unknown unknowns, it may still be possible to explore whether certain effects are occurring — even without knowing causes — by conducting field studies or monitoring small-scale releases.

Paradoxically, seeking to reduce epistemic uncertainty by performing a risk assessment on emerging technologies may require research activities that themselves pose some risk. For example, in the context of synthetic biology, while modelling and assessing risk in closed laboratory situations will lead to answering some safety-related questions, full knowledge of environmental impacts would require some degree of release into the environment. As such, there may be trade-offs between reducing uncertainty and avoiding risk. Decision makers will need to weigh such trade-offs, for example, in permitting field trials or confined field trials of gene drive altered organisms.

3.4 Factors influencing the production of evidence

There are a variety of factors that influence the production of evidence. This discussion is organised around two topics: (i) factors that influence what evidence-related questions are asked (research and development; the economic, political and regulatory contexts; and risk assessment) and (ii) factors that influence how such questions are answered (risk assessment guidelines; who conducts scientific studies). These factors influence what questions are researched, what evidence is produced, and ultimately what evidence is available to inform assessments such as this one and decision making more broadly. Essential to these concerns is the question of who is involved in asking and answering these questions. This section has three objectives: (a) to review the factors that impact what evidence this assessment considers; (b) to describe how the assessment navigated these factors; and (c) to discuss key evidence-related issues that will need to be navigated in future assessments concerning conservation applications of synthetic biology.

3.4.1 Research and development

In research and development, one of the first factors influencing the creation and use of evidence is how a synthetic biology product is designed in terms of desired attributes. For example, in the context of applying synthetic biology to an endangered organism for conservation purposes, this may entail a combination of assessing what traits should change to achieve a conservation goal, what traits should not change, and what ecological outcomes should be achieved. These design goals will impact the creation of evidence because they specify the focus of the studies conducted within the research and development process. This often includes examining potential risks caused by a product and how the design could be changed to minimise them. Due to the significant sway the research and development process has on the final forms technology takes, there have been repeated calls for inclusive engagement to inform this process (Jasanoff, 2003; Macnaghten et al., 2014).

3.4.2 Economic, political and regulatory contexts

Another arena that impacts what questions are asked is the economic, political and regulatory context. First, given the importance of economic profit and societal need in determining technological priorities, political and economic contexts are vital factors influencing what products make it to the research and development stage (see Chapter 6.3). Actors with economic and political power can exert influence over how problems are framed, which influences whether and how synthetic biology applications emerge as potential solutions. The political contestation over framing is thus a key factor influencing the generation of evidence (Bardwell, 1991; Nelson, Andow & Banker, 2009). Second, the regulatory context can also influence how evidence

is created and used, as it often specifies the kinds of studies that need to be completed as part of human health and environmental safety review processes. If there are low levels of trust among communities, stakeholders and regulators, however, product developers may need to conduct a broader set of studies than those required by regulators to create the necessary trust in a product for a successful and supported deployment (Delborne et al., 2018).

3.4.3 Risk assessment

While the process of undertaking this assessment is not a risk assessment in itself, risk assessment is fundamental to the interaction of synthetic biology and biodiversity conservation. As such, this assessment necessarily draws upon the ideas within risk assessment (Chapters 4–6); the formal risk assessment process is also a governance tool for synthetic biology (Section 2.2.1). Ecological risk assessment, or environmental risk assessment, will be an important part of considering the environmental impacts of synthetic biology and gene drive. Ecological risk assessments can take many forms and vary based on the specific regulatory contexts, so the discussion here considers how the basic structure of ecological risk assessment influences the production and use of evidence. Risk assessment, along with risk communication and risk management, are classically defined as the three parts of risk analysis. The ecological risk assessment process embodies a mixture of explicit and implicit decisions impacting the production of evidence concerning the environmental risks of a particular application. Ecological risk assessment contains three major steps: problem formulation, exposure and effects analyses, and risk characterisation (Figure 3.2) (US EPA, 1998). While the specific form that ecological risk assessment takes depends upon the context and stressor in question, the overall steps generally stay the same. There are a host of decisions made across the risk assessment process that influence what questions are asked and determine what studies are called for to adequately assess the potential for harm (Hartley & Kokotovich, 2017).

Problem formulation is where the scope of the assessment and many other foundational decisions are determined, including identifying the stressors, ecological entities and risk hypotheses. *Exposure analysis* is used to assess with what likelihood, under what conditions, and to what extent the stressor will come into contact with the identified ecological entities. *Effects analysis* is used to assess the ecological effects that will result from potential levels of exposure. *Risk characterisation* synthesises the previous stages to assess the risk and address the initial goals of the assessment.

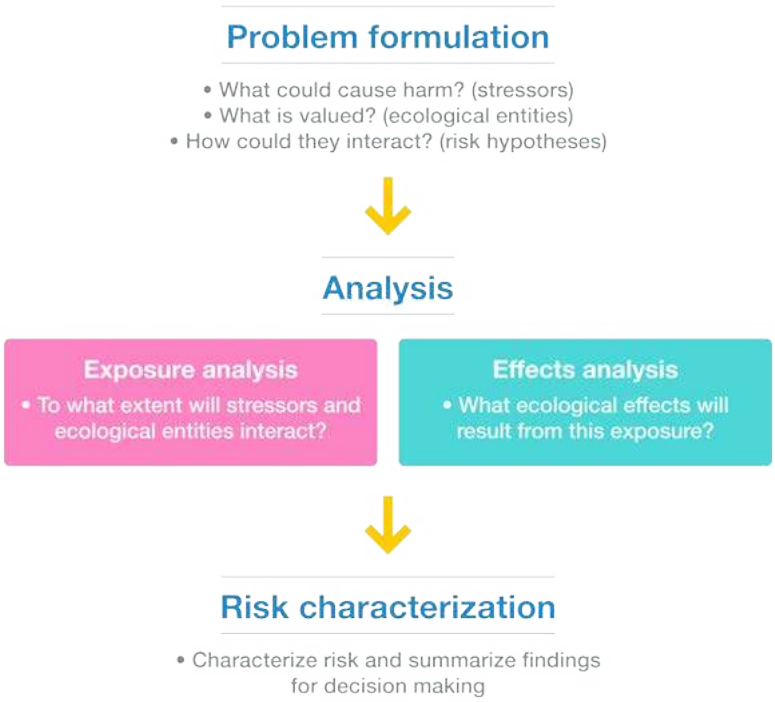

Figure 3.2 Overview of the ecological risk assessment process. Adapted from US EPA, 1998.

Problem formulation is the step in the risk assessment process that most explicitly engages with the factors influencing what questions get asked. The influence of values-based decisions within problem formulation has been recognised and studied (Jensen et al., 2003; Thompson, 2003; Myhr, 2010), and participation from stakeholders and affected parties proposed as a way to transparently and productively incorporate diverse perspectives (Nelson, Andow & Banker, 2009). For example, determining whether changes to ecological entities represent harm, inconsequential change or beneficial change is fundamentally a question of values that may be answered differently based on worldview, perspective or lived experience. To ensure evidence is produced that is decision-relevant and trusted, it is thus important for engagement processes to integrate these value judgements within risk assessment in a deliberative, inclusive and context-appropriate way (NASEM, 2016a; Hartley & Kokotovich, 2017; Thompson, 2018). Engagement around potential applications of synthetic biology or gene drive, therefore, often needs to include local communities and indigenous peoples, who frequently hold different values and perspectives than scientific experts.

It should also be acknowledged that this discussion of risk assessment is relevant for benefits assessment conducted for specific applications. Although the methods for benefits assessment are less developed and agreed upon than for risk assessment, adequately assessing potential benefits will be just as important for informing decision making about whether and how to use applications of synthetic biology and gene drive (NASEM, 2016a). Essential benefits-related questions include, for example, what beneficial impacts are likely to be realised and how will they be distributed? Who gets to define what counts as beneficial? What, if any, non-synthetic biology applications could achieve similar benefits?

3.4.4 Risk assessment guidelines and standards

Similar to the ways that risk assessment can influence what questions get asked, it can also influence how questions get answered; this is also true with regards to formal and informal standards for research processes (Knorr-Cetina, 1999). One part of risk assessment that influences how questions are answered is risk assessment guidelines, the cookbook-like instructions that guide how risk assessments for particular topics are completed (NRC, 1983; Suter II, 2016). While risk assessment guidelines differ based on topic (e.g. for human health, invasive species and genetically engineered plants), different guidelines can also exist for a particular topic. This is significant because these guidelines can call for different processes to be used when conducting studies. For example, differences between two risk assessment guidelines for assessing the non-target impacts of insect-resistant, genetically engineered plants led to a variety of significant differences in how studies are to be completed, including: (1) whether surrogate species or local species are used in tests, (2) whether and when semi-field and field studies are completed, and (3) whether certain indirect effects need to be assessed (Hilbeck et al., 2011; Meyer, 2011; Kokotovich, 2014). These differences can be seen as contributing to different types of "selective ignorance" which result from "the wide range of often subtle research choices or 'value judgments' that lead to the collection of some forms of knowledge rather than others" (Elliott, 2013). As this example illustrates, different guidelines directly impact the form of the resulting evidence and therefore require careful attention.

3.4.5 Who conducts studies

Similar impacts on knowledge production can emerge as a result of who conducts studies that feed into risk assessment processes. First, the subtle research choices that are part of conducting scientific studies may be influenced by recognised or unrecognised assumptions and biases about a product's safety (Krimsky, 2013). Therefore, it is vital to be aware of this potential and to remove potential conflicts of interest from the conduct of research. Second, differences across disciplines can alter how questions are answered – an ecologist will likely design and conduct studies differently than a toxicologist. Thus, disciplinary, institutional and personal affiliations all combine to influence the production of knowledge in the context of risk assessments. This phenomenon can be managed to some degree with explicit risk assessment

guidelines, commitments to transparency and the avoidance of perceived or real conflicts of interest.

3.4.6 Situating this assessment

This assessment relates to evidence in two ways: it is at once informed by evidence and also will serve as evidence. Throughout, the chapters draw upon and synthesise existing scientific studies, and are therefore based upon existing evidence. At the same time, this assessment will serve as an input into the IUCN policy process, and in that way it *is* evidence.

One might ask, then, what this chapter on evidence is *evidence* for. First, while it falls outside of the scope of this assessment to conduct full-scale risk and benefit assessments on specific applications, this chapter affirms that risk and benefit assessments will provide a vital set of evidence to inform decision making on specific products and applications on a case-by-case basis. What form risk and benefit assessment guidelines take and who is involved in conducting these assessments will be of consequence to the production and synthesis of evidence for decision making. The case-by-case nature of this decision making is key. The concern has been raised that conservation uses of synthetic biology will serve as a smokescreen for detrimental uses. That is, applications of synthetic biology that seem beneficial for the environment will lead societal actors, government regulators and the public more broadly to turn an uncritical eye towards future, more questionable synthetic biology applications such as those involving military-related ends or the corporate control over agriculture. However, the fact that one application may be beneficial in a certain social, political, economic and ecological context does not imply that the same technology would be beneficial in another context, and does not imply that other applications are more likely to be beneficial. Furthermore, different applications require different assessments, even if some knowledge is transferable. Polarised thinking that bundles all synthetic biology applications together for summary judgement, for or against, masks this complexity in favour of highly charged politics that fails to notice when different applications of synthetic biology could be beneficial, detrimental or a mix of both. Thus, this assessment should not be read as a judgement – positive or negative – on all synthetic biology or even all conservation applications of synthetic biology. Rather, it serves as an initial discussion of factors that will need to be considered in case-by-case decision making by the full range of appropriate stakeholders, operating with free access to all information, and informed by the framework of the precautionary principle.

Second, while it also falls outside of the scope of this assessment to suggest whether and how research and development on synthetic biology and gene drive *should* advance, this chapter emphasises how the evidence that will often take centre stage in such debates will depend not just on a narrow definition of scientific rigour, but rather on the way that multiple perspectives and values create the context for knowledge production. Important questions include how successful products are defined, what values determine the formulation of problems, how engagement integrates multiple perspectives in risk assessment processes, and who is trusted to produce credible knowledge.

In the spirit of reflexivity, this chapter concludes with a reflection on such questions with regards to the assessment as a whole. First, in terms of context and scope, the priorities of the IUCN membership helped focus this assessment uniquely on the conservation implications of potential synthetic biology applications, including engineered gene drive systems. These priorities also helped determine the analytical framework used in this assessment to analyse potential applications of synthetic biology (see Chapter 4).

Second, the selection of authors for this assessment is clearly consequential. Authors have primary expertise in a diversity of areas including synthetic biology, engineered gene drive systems, natural science, social science, conservation management, governance and law. A key aspect of this assessment's scope involved an examination of the potential synthetic biology and engineered gene drive applications relevant to the conservation and sustainable use of biodiversity. To do this rigorously required engaging with those who are knowledgeable about the applications, such as those who have been involved in its development. Readers will notice that the authors of the case studies used in Chapters 5 and 6 are developers or

researchers closely tied to these applications. Many of these case study authors, due to their involvement in research and innovation processes, demonstrate enthusiasm for the potential of these applications to impact the world in beneficial ways. To provide balance, the assessment lead authors ensured that the discussion explored potential detrimental impacts on the conservation and sustainable use of biodiversity, as well as broader social, economic, cultural and ethical considerations, as mandated by WCC-2016-Res-086. Thus, Chapter 2 explores issues surrounding governance, broadly defined; this Chapter 3 offers a critical and reflexive view of the production and use of evidence; and the case studies in Chapters 5 and 6 specifically address each of the following questions:

- What is the conservation issue to be addressed?
- What are the existing interventions and their limitations?
- What is the synthetic biology intervention under consideration?
- What are the potential conservation benefits of the synthetic biology approach?
- What are the potential adverse effects and limitations of the approach?
- What are the relevant social, economic and cultural considerations?

Yet, even with attention to this balance of questions and the disciplinary diversity of the assessment authors, not all potential perspectives were captured in the assessment. This shortcoming was at least partially rectified through the process of open peer review, which generated 742 comments and critiques from persons across the globe that informed revisions to this final version of the assessment.

This chapter thus concludes with the observation that this assessment is not – and cannot be – perfectly objective, unbiased and comprehensive. Instead, this report as a whole offers evidence and frameworks for analysis with the aim of informing future deliberations, within IUCN and more broadly, about the responsible innovation and governance of synthetic biology and engineered gene drive systems.

4 .
Analytical framework for assessment of synthetic biology and biodiversity conservation

Delphine Thizy, Thomas M. Brooks, Nicholas B.W. Macfarlane, Aroha Te Pareake Mead

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This assessment analyses synthetic biology and engineered gene drive and their potential impacts on biodiversity conservation in two ways: in general terms, summarising across the available evidence for the different potential types of application, and through case studies. This chapter focuses on the second of these types of evidence, the case studies.

4.1 Role of the case studies

The case studies in the chapters that follow provide examples of potential applications of synthetic biology and engineered gene drive to conservation. The goal of the examples is to supplement the broader analysis of the evidence regarding such applications withmore focused discussions of potential benefits and harms.

The primary technical section of this assessment (Chapters 4–6) follows the two main categories of synthetic biology applications that can potentially impact biodiversity conservation:

- synthetic biology applications intended to have a conservation benefit (Chapter 5); and
- synthetic biology applications that have a different primary aim but could also have impacts on conservation goals (Chapter 6).

4.2 Selection process for case studies

The potential synthetic biology applications in conservation and across sectors of society are too broad, and the speed at which they are currently being developed too rapid, to describe all of them in this document.

The Technical Subgroup is committed to the principle of *inclusivity*. Accordingly, the Technical Subgroup selected case studies in a way designed to present the breadth of issues that synthetic biology and engineered gene drive applications are trying to address — with respect to both conservation objectives and to diverse threats.Therefore, Chapter 5 is structured using two key conservation objectives: reducing threats to species; and improving species, community and ecosystem resilience to threats.

When selecting the case studies, the Technical Subgroup carried out a horizon scanning of proposed applications to identify those that were already in development, as well as those frequently mentioned, either through publication, personal familiarity, or because they raise particular concerns by civil society or within parts of the conservation community. Only case studies for which there was adequate evidence of potential impacts were included. There is one additional box on chytridiomycosis in amphibians, for which there is no synthetic biology solution under development, but it is included because of its significant potential impact on biological diversity. There were initially 14 case studies considered for Chapters 5 and 6, but some of them were subsequently dropped because of lack of sufficient published evidence.

Case studies in Chapter 6 draw on applications from the broad categories of product replacement and pest management. The chapter also briefly discusses potential agricultural applications. There are clearly other categories in which synthetic biology interventions might indirectly affect conservation (e.g. in human health) but at this stage discussion of most of these would be speculative rather than evidence-based. The level of available knowledge differs amongst applications: for some, research is quite advanced and scientists participating in these developments were involved in drafting the case studies. For others, the Technical Subgroup members had to rely on publiclyavailable evidence.

The case studies represent a diversity of applications and the level of data and evidence available for them varies (Table 4.1). This is one of the primary challenges for Chapters 5 and 6, and the authors have highlighted the related uncertainties pointing to areas where additional data are needed.

Case studies also vary in their scale and specificity. Some consider application to particular regions (e.g. preventing avian malaria impacts to Hawaiian birds), while others, frequently only speculative or at earlier stages of development, are more broadly

looking at the opportunity of using synthetic biology and engineered gene drive to address particular conservation goals (e.g. potential approaches to control or eradicate rodentsimpacting island biodiversity).

Table 4.1 Characteristics of the case studies presented in Chapters 5 and 6.

Case study	Chapter	Context and scale	Stage of development
1. **Eradicating invasive rodents from islands**	5	Rodents impact island biodiversity globally	Technical development underway, but laboratory proof of concept still to be achieved; Social and policy engagement underway.
2. **Controlling invasive mosquitoes to prevent bird extinctions in Hawaii**	5	Vectors of disease impacting birds in Hawaii	Technical development underway, but laboratory proof of concept still to be achieved; Analysis of socio-economic and cultural considerations hasn't yet been carried out.
3. **Synthetic biology to address conservation threats to black-footed ferrets**	5	Disease impacting black-footed ferrets in North America	Technical approach speculative; could have a significant economic impact because of the indirect impact on grassland/ prairie ecosystem recovery.
4. **Transgenic American chestnut for potential forest restoration**	5	Disease impacting American chestnut in North America	Trees potentially ready for field trials; Specific research on socio-economic and cultural considerations should be carried out to identify specifically what the benefits or adverse effects could be.
5. **Corals and adaptation to climate change/acidification**	5	Ocean warming impacting coral globally	Technology development in early stages; Specific socio-economic and cultural consideration assessment would need to consider a particular intervention in a given ecosystem and context.
6. **Horseshoe crab replacement for *Limulus Amebocyte Lysate* test**	6	Four Asian and North American species threatened by overuse in biomedical industry	Recombinant assay available since 2003; factors such as uncertainty over efficacy, regulation, availability and industry inertia have limited its adoption.
7. **Gene drive approach for malaria vector suppression in Africa**	6	Target transmission of malaria parasite by suppressing population of *Anopheles* mosquitoes in Africa	Technical development underway, but laboratory proof of concept still to be achieved; Analysis of socio-economic and cultural considerations hasn't yet been carried out.
8. **Addressing honeybee colony collapse**	6	Colony Collapse Disorder associated with widespread loss of managed honeybee colonies	Technical approach speculative; significant loss of pollinators can have a large impact on biodiversity and ecosystem services in natural landscapes.

4.3 Analytical framework for the case studies

IUCN Resolution WCC-2016-Res-086 calls for an examination of "the organisms, components and products resulting from synthetic biology techniques and the impacts of their production and use, which may be beneficial or detrimental to the conservation and sustainable use of biological diversity and associated social, economic, cultural

and ethical consideration," and "the implications of engineered gene drives and related techniques and their potential impacts on the conservation and sustainable use of biological diversity as well as equitable sharing of benefits arising from genetic resources." The case studies presented in Chapters 5 and 6, commissioned for this assessment, are centred around six questions, to provide the necessary context and provide examples of potential impacts:

- What is the conservation issue to be addressed?
- What are the existing interventions and their limitations?
- What is the synthetic biology intervention under consideration?
- What are the potential conservation benefits of the synthetic biology approach?
- What are the potential adverse effects and limitations of the approach?
- What are the relevant social, economic and cultural considerations?

4.3.1 Conservation issue

The case studies present a range of conservation issues that might be addressed through synthetic biology tools and techniques. For Chapter 5, these issues are directly related to conservation goals, while for Chapter 6 the issues are not directed at achieving conservation goals but are designed for other objectives (e.g. agriculture, human health, product replacement) but might have secondary impacts on conservation.

The authors of these case studies were selected on the basis of their expertise and familiarity with conservation issues and the potential synthetic biology applications. They are uniquely suited to describe the situation in a way that will foster deeper understanding of how synthetic biology and conservation may intersect. In most cases, the authors also have a strong interest in investigating the feasibility of the application, so they are not entirely neutral observers. The overall chapter authors reviewed the case studies (see *Principle-based assessment* section below) and provide the necessary context for the case studies in the accompanying text.

4.3.2 Existing interventions and limits

While current conservation actions are yielding substantial positive impacts on biodiversity (Hoffmann et al., 2010), overall they are still falling well short of delivering on societal expectations and intergovernmental commitments to halt extinctions, prevent conversion of natural ecosystems, and maintain genetic diversity (Tittensor et al., 2014). In some cases, appropriate conservation tools exist, but the extent of their deployment is insufficient to mitigate the threat. For example, protected areas can be effective in safeguarding key biodiversity areas (Butchart et al., 2012) , even though many protected areas are not located in the most important places (Venter et al., 2018) and are insufficient to mitigate external threats like climate change (Bruno et al., 2018).

The insufficient deployment of conservation tools is often due to a lack of resources, vested interests that oppose conservation, and other limitations that constrain taking successful conservation actions to the necessary scale. In addition, there are some gaps between actions and impacts because conservation tools simply do not exist to mitigate certain threats, for example, chytrid fungal disease in amphibians (Section 5.3.1).

The case studies attempt to frame the potential benefits of proposed synthetic biology and engineered gene drive approaches in light of current, more conventional conservation interventions (in other words, the "counterfactual" (Ferraro & Pattanayak, 2006)). As such the current interventions and their potential limits are briefly analysed.

4.3.3 Synthetic biology description

Each case study describes how synthetic biology may be used to address the identified challenge. As not all approaches are equally advanced, the descriptions range from theoretical ideas that still need to be demonstrated, to applications that have already been extensively researched and have yielded proof of principle (showing evidence that the concept can work as anticipated in the

laboratory) to a few cases where researchers have evidence that their technology is ready to make regulatory applications for small-scale field testing.

The case studies briefly describe the proposed synthetic biology approach, and what the application intends to achieve. In most cases, the case studies do not provide the technical details of these applications; references are provided to scientific papers, and Appendix 3 (www.iucn.org/synbio) presents the technical details. The scale or specificity of the approach is also included in the technology description. To the fullest extent possible, the case studies describe the feasibility of the proposed approach and its current level of progress.

4.3.4 Potential conservation benefits

This assessment is not a benefit assessment, but rather provides initial indications of potential conservation benefits that might warrant further considerations in future case-by-case benefit assessments. This section looks at the extent to which synthetic biology applications could potentially complement or even replace existing interventions or address some of the limits of those interventions. This section also points to knowledge gaps that are common for technologies in early stages of development.

4.3.5 Potential adverse effects and limitations

This assessment is not a risk assessment, but rather provides initial indications of potential adverse effects that might warrant further considerations in future risk assessments. Concerns raised by stakeholders are also presented when known from literature or other published information. Risk assessment is a complex and thorough process for any technology (Section 3.4.3). Existing risk assessment frameworks, some of which are embedded in regulation, and information on past exercises (for instance from the Living Modified Organism experience) are also available (Section 2.2.1). A full-fledged risk assessment requires, among other things, consideration of the specific characteristics of the organisms, as well as of the receiving environment (UN CBD, 2000).

4.3.6 Social, economic and cultural considerations

All significant conservation actions have consequences for human economy and cultures, some positive and some negative. Conservation has often had particularly significant impacts, positive and negative, on indigenous, and other peoples due to their greater reliance on goods and services from the natural environment (Garnett et al., 2018).

Socio-economic and cultural considerations are important in assessments by IUCN and other institutions, as well as decision-making frameworks for technologies (UN CBD, 2000). Conner (2016) proposes a rigorous approach for full evaluation of socio-economic dimensions that can analyse both the potential opportunities and adverse impacts. However, similar to a risk assessment, the assessment of socio-economic and cultural considerations requires specific information about the technology proposed and the receiving society, economy and culture. Therefore Chapters 4 and 5 identify areas where socio-economic and cultural considerations would need to be taken into consideration according to existing evidence — when available — and will warrant further studies and/or consultation, usually on a case-by-case basis. Whenever possible, the socio-economic and cultural considerations for the proposed synthetic biology approach should be weighed against those of more conventional interventions.

4.3.7 Principle-based assessment

Scholars studying responsible research and innovation emphasise the importance of key principles to inform the governance of emerging technologies (Chapter 2.1). Recognising the need for a broad and inclusive process, the Technical Subgroup charged with completing this assessment developed a set of principles that have guided its work (see Front Matter). The underlying principles of *objectivity* and *robustness* adopted for this process have been rigorously applied to each case study. The process of writing these case studies has been iterative. Case study authors were asked to present their text based on the framework and the

Technical Subgroup members reviewed the objectivity and robustness of content. Drawing from the principle of *objectivity*, the Technical Subgroup then ensured that the case studies were objective; this is, they were not written from an advocacy perspective but that statements were based on available evidence. The reviewers also ensured that the cases were robust; that is to ensure that the strength of statements made was justifiable. Case studies are presented in boxes, with context provided in the accompanying text of Chapters 5 and 6 to fulfil the mandate outlined in the Resolution.

Many case studies present applications that are still in the early days of development and testing, so a high degree of uncertainty remains about what the final technology could be, as well as the exact potential benefits, adverse impacts, and social, economic and cultural considerations. It is essential to recognise uncertainty when presenting these case studies (Section 3.3), and when possible to identify some of the knowledge gaps that might need to be further evaluated for decision making on potential technology application. Although not within the scope of this assessment, rigorous, context specific, case-by-case risk assessments need to be completed for any future proposed synthetic biology and engineered gene drive applications. The discussion of potential adverse effects included within these case studies may help inform these formal risk assessments, but they are in no way substitutes for them. Oliver (2018) demonstrates how uncertainties — often confused with risks — can shape public acceptance and decision making. It is thus important to recognise the level of uncertainty in the case studies.

5.
Synthetic biology applications intended for conservation benefit

Daniel M. Tompkins, Elizabeth L. Bennett, Hilde Eggermont

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5.1 Overview

Biodiversity decline has dramatic ecosystem-wide consequences for how species interact, how communities are organised, and the ability of ecosystems to provide services such as nutrient cycling and carbon sequestration. The loss of biodiversity is at least equal to climate change as a driver of ecosystem change (Hooper et al., 2012). About 50 per cent of the Earth's land has been converted to cropland, rangeland and urban areas, with net natural forest area declining annually by 65,000 km^2 between 2010 and 2015 (FAO, 2017).

In the past two decades, many global, regional and national policies and legislations promoting biodiversity conservation have been adopted or expanded (Section 1.8). Some successes have been achieved (Sodhi et al., 2011); examples include the recovery of great whale populations globally (Gales, 2011), and the more local improving conservation status of the giant panda (Swaisgood, Wang & Weif, 2016; Xu et al., 2017) and Arabian oryx (IUCN SSC Antelope Specialist Group, 2013; Barichievy et al., 2018). However, biodiversity continues to decline globally, with an estimated 25 per cent, 13 per cent and 41 per cent of all mammals, birds and amphibians respectively threatened with extinction in 2017, up from 19 per cent, 10 per cent and 2 per cent respectively in 1996/98 (IUCN, 2017).

The conservation of biodiversity requires the continued application of proven approaches; e.g. a greater proportion of the planet being designated as protected areas and managed effectively (Jones et al., 2018), and a step-up in the management of invasive alien species (IUCN, 2016). However, past experience has shown that scaling these efforts up to the level necessary to reverse the declines in biodiversity and allow for

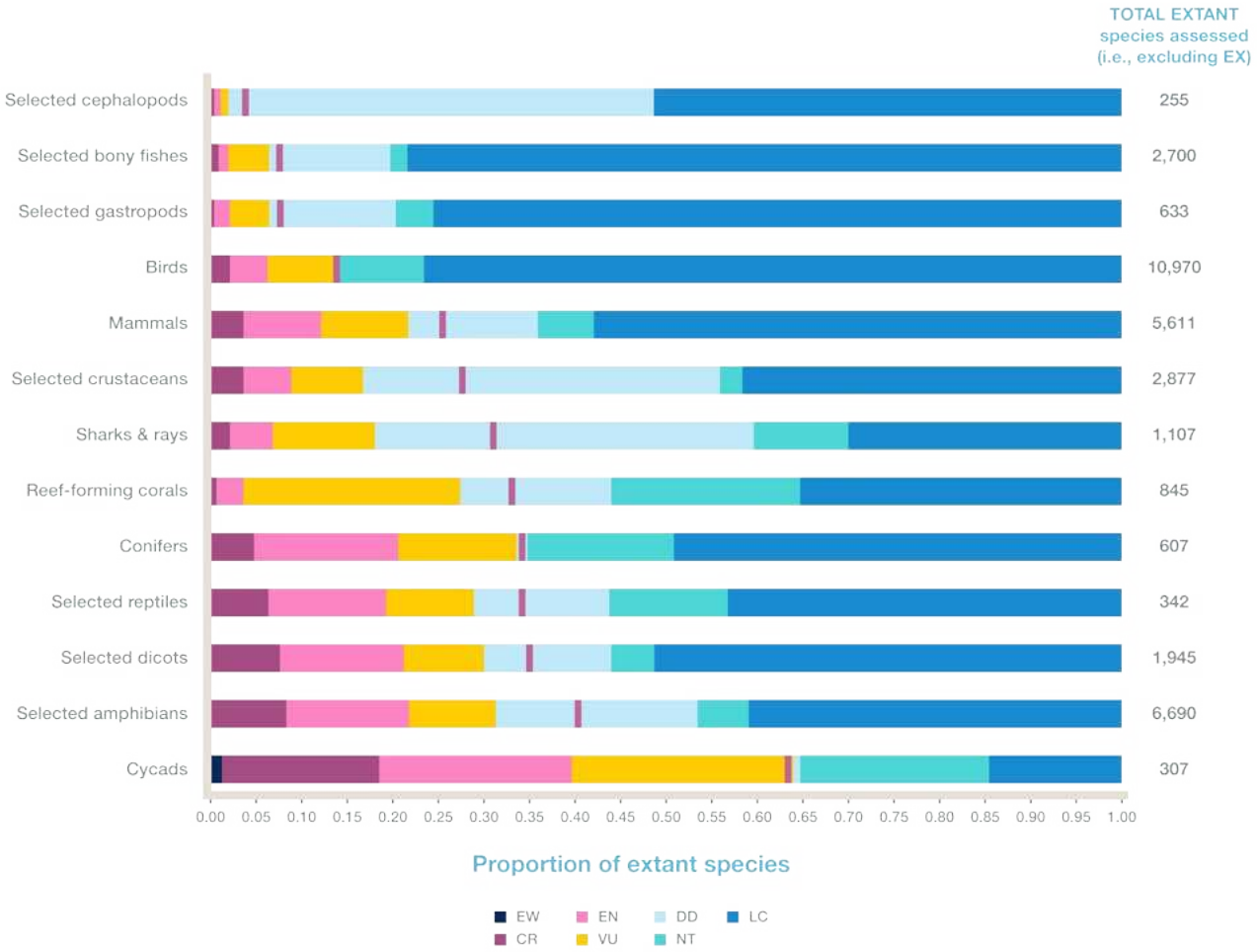


Figure 5.1 The proportion of extant (ie., excluding extinct) species in The IUCN Red List of Threatened Species. Version 2019-1 assessed in each category for the more comprehensively assessed groups. The numbers to the right of each bar represent the total number of extant species assessed for each group. EW - Extinct in the Wild, CR - Critically Endangered, EN - Endangered, VU - Vulnerable, NT - Near Threatened, DD - Data Deficient, LC - Least Concern. Adapted from IUCN Red List of Threatened Species Summary Statistics. http://www.iucnredlist.org

recovery will continue to be a major challenge using current approaches, given the costs and the seemingly intractable nature of some of the threats (Veitch & Clout, 2002; Glen et al., 2013). Certain synthetic biology applications, if appropriately designed and targeted, may have potential for enhancing biodiversity conservation, while others could potentially damage it (Redford et al., 2014; Piaggio et al., 2017).

This chapter explores how engineered gene drives and synthetic biology organisms, applications and products might directly benefit or impact conservation through their use for the purpose of conservation management. It thus focuses specifically on situations where the intended use of synthetic biology is to achieve conservation goals or protect conservation values. It explores the potential positive conservation outcomes from such applications and details important considerations, while also recognising that many situations, if not managed appropriately, could potentially also have negative impacts on conservation. The chapter will review potential applications of synthetic biology and engineered gene drives that could reduce threats to species, and improve species, community and ecosystem resilience to threats. It will introduce and draw upon specific case studies that use the framework outlined in Chapter 4 to illustrate the direct conservation impacts, both positive and negative, and the potential benefits and adverse effects associated with some of the potential applications of synthetic biology.

5.2 Mitigation of threats

5.2.1 Tackling invasive alien species

The increasing global complexity of transportation systems on land, air and sea has broken down the natural barriers to species movements formed by rivers, oceans and mountains, the barriers that isolated populations and allowed species diversity to evolve and be maintained (DiCastri, 1989; Meyerson & Mooney, 2007). As a result, invasive alien species effects on native wildlife and ecosystems are immense (IUCN, 2000) being the second biggest driver of species extinction (Clavero & García-Berthou, 2005; Bellard, Cassey & Blackburn, 2016) and having large negative impacts on ecosystem function (Pejchar & Mooney, 2009; Ehrenfeld, 2010). Invasive alien species also cause multiple other costs globally; for example, they cause huge infrastructure damage (Scalera et al., 2012; IASC, 2016), and agricultural losses in Australia due to invasive alien species were estimated to be worth an average of A$ 620 million per annum over five years (Gong et al., 2009).

Invasive alien species are found in all taxonomic groups, from fungi and bacteria to mosses, higher plants, invertebrates and vertebrates (Lowe *et al.*, 2000), and their impacts can be exacerbated by habitat disturbance and climate change (Early *et al.*, 2016). Of 170 animal extinctions for which the causes of extinction are known, 20 per cent and 54 per cent are solely and partly due to invasive alien species respectively (Clavero and García-Berthou, 2005). A total of 1352 mammal, bird, reptile and amphibian species worldwide classified as threatened (i.e. in the IUCN Red List, Vulnerable, Endangered or Critical categories) are primarily imperilled by invasive alien species impacts (Bellard, Genovesi and Jeschke, 2016). The number is particularly high for amphibians (*N* = 565; ~8 per cent of amphibian species) and birds (*N* = 443; ~5 per cent), compared with mammals (*N* = 183; ~3 per cent) and reptiles (*N* = 161; ~2 per cent).

Invasive alien species are the primary driver of species extinctions on islands (Doherty et al., 2016; Spatz et al., 2017). Islands make up 5.3 per cent of the Earth's land area, yet maintain an estimated 19 per cent of bird species, 17 per cent of rodents and 17 per cent of flowering plants (Tershy et al., 2015). They are also home to invertebrate assemblages with frequently high levels of endemicity and often performing key ecosystem functions (St Clair, 2011). Species diversity is disproportionately threatened on islands in relation to the islands' proportion of both global land area and species, with 37 per cent of all critically endangered species being confined to islands. Sixty-one per cent of all extinctions within the last 500 years have been island species, and invasive alien species are one of the most important threats to remaining insular diversity. For vertebrates, seabirds are especially vulnerable since most species are obligate island breeders where their colonial ground-

breeding behaviour has evolved in isolation from terrestrial predators (Schreiber & Burger, 2001). For invertebrates, large-bodied species are particularly threatened by invasive rodents (St Clair, 2011).

Non-native diseases, frequently vectored by non-native animals, also have had and continue to have large impacts on animal, plant and human health, impacting biodiversity and other values (Crowl et al., 2008; Hulme, 2014; Tompkins et al., 2015). For example, avian malaria, vectored by non-native mosquitoes, is the primary cause of endangerment and extinction of endemic Hawaiian honeycreepers, among the most critically endangered birds globally (Liao et al., 2017), with impacts also in regions such as the Galapagos Islands (Wikelski et al., 2004). Similarly, the spread of chytrid fungus threatening amphibians globally (Fisher, Garner & Walker, 2009) is considered to have been facilitated by the introduction of non-native species (Fisher & Garner, 2007). Such issues can potentially be addressed by managing either threatened hosts (e.g. management to increase their resilience to disease) or any vectors of disease (e.g. by reducing their populations or their vector competence).

Synthetic biology offers potential novel approaches to managing invasive alien species, but as with any management approach there are also potential adverse effects (Harvey-Samuel, Ant & Alphey, 2017; Ricciardi et al., 2017). The applicability of different actions for invasive alien species management tends to vary across scale, with the success of actions frequently being limited as spatial scale increases (Veitch & Clout, 2002; Glen et al., 2013). In addition to the management of established 'legacy' invasive alien species that are currently impacting biodiversity, synthetic biology and engineered gene drive also offer novel potential approaches for rapid response and eradication of new invasive alien species incursions. In such contexts, synthetic biology application may be more feasible and have less potential adverse effects due to management efforts being more tactical, targeted and at a smaller scale. The benefit and adverse effect profiles of applying potential synthetic biology approaches to the management of invasive alien species will thus likely vary with both application scale, context and targeted species or population.

5.2.1.1 Potential synthetic biology applications: Management of invasive vertebrates

Especially damaging invasive alien species include cane toads, rodents, pigs, goats, carp and crayfish, and mammalian predators such as feral cats, foxes, raccoons, stoats and mongooses (Moro et al., 2018). Invasive mammals are the main cause of animal extinctions on islands, both by direct predation – especially of birds and their eggs – and also destruction of native habitat (Doherty et al., 2016; Spatz et al., 2017). Feral cats on islands are responsible for at least 14 per cent of global bird, mammal and reptile extinctions, and are the principal threat to almost 8 per cent of critically endangered birds (mainly seabirds), mammals and reptiles (Medina et al., 2011).

Eradicating invasive mammals has been attempted on more than 700 islands globally; at least 107 highly threatened birds, mammals and reptiles on the IUCN Red List of Threatened Species (6 per cent of highly threatened species) have probably benefited from invasive mammal eradication on islands (Jones et al., 2016). Some species are seemingly easier to eradicate than others; goats have been successfully eradicated from 120 islands worldwide using a combination of approaches (Campbell & Donlan, 2005). Rodents have been eradicated successfully on 73 per cent of the 387 islands where rodenticide programmes have been deployed; however, for many other islands where invasive rodent eradication would benefit threatened species it is not possible to use rodenticides at this time due to social and biological barriers (Howald et al., 2007; Campbell et al., 2015). Feral cat eradication had been achieved on at least 83 islands worldwide, including 11 large islands over 2000 hectares (Parkes et al., 2014), but remains challenging particularly on islands with significant human presence (Nogales et al., 2004).

Current control techniques for invasive alien mammal species typically consist of integrated chemical and physical management practices (e.g. poison baiting combined with fencing), direct intervention (e.g. shooting, trapping) and biological control with natural enemies (Eason et al., 2017). Eradication programmes can be costly, particularly over larger

areas, and have varying levels of efficacy (Bomford & O'Brien, 1995; Courchamp et al., 2003); eradicating invasive alien species entirely is challenging, and the results of local eradication are often short-term if sources for reinvasion persist (Myers et al., 2000). Impacts on non-target species are also a concern; for example, poison deployment requires extensive planning and caution to minimise mortalities of non-target species (Pitt et al., 2015; Novak, Maloney & Phelan, 2018). Poison baits aimed at invasive alien species have caused declines of non-target species that eat them, although populations in most cases then recover once the invasive alien species have been eradicated (Jones et al., 2016). Biological control of invasive alien mammal species offers target species-specificity and landscape scale applicability (due to its self-disseminating nature), for which there have been some population suppression successes (e.g. biological control of rabbits in Australia; Cooke et al., 2013); but evolution of resistance generally necessitates periodic release of novel control agent strains or species (Cox et al., 2013) and there are societal concerns over non-target population impacts (e.g. to domestic rabbits). There is also increasing concern with regards to animal welfare about the impacts of current control techniques for invasive alien species management, particularly when applied to invasive mammals (Littin et al., 2004; Warburton et al., 2012). These constraints are causing scientists and managers to seek additional tools that are more species-specific, economical, self-sustaining, and with lower animal welfare impacts; however, there are currently no widely-applicable alternative solutions (Campbell et al., 2015).

For potential synthetic biology application to vertebrate invasive alien species, attention is currently focusing on engineered gene drive systems (Chapter 1.4) with the potential for self-dissemination through populations over generations (Campbell et al., 2015; Piaggio et al., 2017). This is because, with the exception of approaches based on traditional biocontrol, non-gene-drive control is generally not self-disseminating and thus logistically more challenging to employ at a landscape scale (Moro et al., 2018). While researchers can envisage non-gene-drive solutions for some vertebrate pests, such as cane toad, bighead carp and sea lampreys (Harvey-Samuel, Ant & Alphey, 2017), such approaches have more potential hurdles to overcome (linked to the large numbers of organisms needing to be reared and released) than approaches with more potential for self-dissemination such as engineered gene drive systems (Appendix 3 (www.iucn.org/synbio)).

All engineered gene drive systems are currently only theoretical for application to vertebrates (Appendix 3 (www.iucn.org/synbio)). Functional engineered gene drive mechanisms are not yet developed in such species (Grunwald et al., 2018). There is also the challenge of identifying genetic manipulations that cause the desired population-scale effects when spread by drive mechanisms (Gemmell & Tompkins, 2017). These could potentially be of multiple forms, such as gene cargos (genes that drive mechanisms are used to increase the frequency of) or endogenous gene knock-outs (i.e. drive mechanisms spreading loss of function) that cause population decline when spread (Burt, 2003). With the relative ease of applying the CRISPR-Cas9 toolkit for gene editing, the identification of suitable genetic manipulations to spread with engineered drive mechanisms is likely the bigger hurdle, and there is no evidence yet that it can be overcome for vertebrate pest targets.

Engineered gene drive approaches to invasive alien species management will also be generally less applicable to invasive vertebrates than other invasive alien species taxa, due to gene spread through populations taking longer to occur in species with longer generation times. There are also practical issues that may limit gene drive efficacy when applied in the field, such as reproductive behaviour (Gemmell & Tompkins, 2017), spatial and temporal heterogeneity in populations and landscapes (Deredec, Burt & Godfray, 2008) and evolution of resistance (Unckless, Clark & Messer, 2016; Champer et al., 2018). Thus, such approaches, while offering potential for application to vertebrate pests, have several developmental hurdles to overcome. Given the relationship to generation time noted above, and the role of the mouse as a model species for genetic studies, should engineered gene drive population suppression or eradication for vertebrate invasive alien species management be achievable it will most likely be first developed for rodents.

Case study 1:

Eradicating invasive rodents from islands

Nick Holmes, Karl Campbell

Issue

Rodents remain one of the most widespread invasive species, estimated to occur on 80 per cent of the world's island groups (Atkinson, 1985), where they contribute to species decline and extinction via mechanisms including predation and disruptive habitat modification. The eradication of invasive rodents has been applied on more than 500 islands globally (Russell & Holmes, 2015), with demonstrable conservation benefit (Jones et al., 2016).

Existing interventions and limitations

The current toolbox for insular rodent eradications relies on the sufficient application of anticoagulant rodenticides into every potential rodent territory on an island (Broome et al., 2014). Despite significant advances in the use of these toxicants over recent decades, in many situations these eradication methods are extremely challenging or unfeasible. These include islands with significant human populations, stakeholder communities adverse to the method, co-occurrence of livestock and domestic animals, or potential negative impacts to native species (Campbell et al., 2015).

Synthetic biology and engineered gene drive description

Synthetic biology presents potential solutions to overcome these challenges by using, among other approaches, engineered gene drive systems to bias gene inheritance throughout a population to drive it to local extinction, for example by distorting the sex ratio (Webber, Raghu & Edwards, 2015). In such cases, the drive mechanism employed would need to be strong enough to overcome any selective disadvantage incurred by the individuals carrying the genetic manipulation for it to spread. The potential benefits include species specificity, reduced toxicant use, more humane (non-lethal) approaches and expanded application on human inhabited islands (Campbell et al., 2015). This represents a potentially transformative advance for the island restoration field not readily achievable with current technology.

Potential adverse effects and limitations

Three general potential adverse effects are evident in considering synthetic biology for invasive rodents. First is the concern of direct effects on the biology and ecology of non-target species, and associated community and ecosystem knock-on effects, due to genetic changes spreading to them from targeted species. Since animals are largely unaffected by horizontal gene transfer and thus gene transfer generally only occurs through sexual reproduction (Andersson, 2005), this is a minimal concern for islands where no related species occur (Campbell et al., 2015). In addition, multiple genes are generally needed for phenotypic change, and these vary from species to species (Johnson et al., 2016).

Second is the concern of effects on non-target populations of the same species, and associated community and ecosystem knock-on effects, due to gene modified organisms moving beyond the target population; i.e. they swim or are transported beyond the target site by human activity. The likelihood of such occurring is logically reduced if the target population is restricted to one or more isolated islands rather than being closer to

— **House mouse (*Mus musculus*)** (Rudmer Zwerver / Shutterstock.com)

other land-masses. Notably, rodent incursions to islands have been effectively managed using biosecurity policy and protocols (Russel et al., 2008), offering confidence in the ability to prevent rodent movement *away* from islands. Third is the concern that rodent elimination could have adverse effects on food webs or impact ecosystem processes; however, this could occur for eradications using either traditional toxicant or synthetic biology methods, and consideration of such effects is an existing recommendation for management (Zavaleta et al., 2001).

Social and regulatory acceptability are as significant as technical factors, and these three components are prerequisites for any potential field trial or future release. Social acceptability will be strongly influenced by the public's perceptions of the need for action, potential efficacy of the technology, potential benefits and adverse effects, and how these inter-relate with socio-economic and cultural factors. Regulatory acceptability will depend upon the specific country, state, local regulations and case-by-case assessments.

Technical uncertainties in deploying genetic biocontrol to eradicate invasive rodents from islands include the engineering of modified rodents, competitiveness of modified rodents in wild populations, and potential resistance to engineered gene drive systems over multiple generations (and hence the ability to achieve 100 per cent gene transfer throughout the population to achieve eradication). Research needed to reduce these technical uncertainties, allowing adverse effects to be minimised and potential gains to be maximised, includes advancing knowledge of genome engineering, mating success of engineered and wild rodents, mechanisms to contain engineered gene drive systems locally or temporally (Dhole et al., 2018) and delivery strategies. Such knowledge will be necessary for any field trial proposal to be effectively evaluated.

There is yet no consensus on what type of field sites may be best for trialling genetic biocontrol. At this stage, potential trial sites need to be considered on a case-by-case basis. Key technical factors on which such consideration should

be based, for potential application to invasive rodents on islands, would include characteristics of the target population, the local ecosystem, the characteristics of the modification introduced, the potential for off-island dispersal, the ability to conduct comprehensive monitoring and the ability to shut down trials (e.g. with traditional rodenticide methods) should such a step be required.

Technologies to engineer mice, and more recently rats, are well established and have been used for several decades for biomedical applications. The genetic approaches for eradicating or reducing the impact of invasive rodents are still in their infancy; the timeline to develop a comprehensive field trial proposal is estimated to be a decade (http://www.geneticbiocontrol.org/). Urgency exists because the motivation for developing new tools – extinctions and endangerment on islands – continues (Doherty et al., 2016).

Socio-economic and cultural considerations

Although the situation will differ depending on the island considered, some potential areas for impacts of rodent eradication using synthetic biology approaches on socio-economic and cultural considerations can be identified: (i) perception of likely effectiveness of the method; (ii) acceptability of genetic modification as interpreted by cultures and belief systems at a particular site; (iii) perceptions of, and likely positive and negative impacts to natural resources and culturally significant species; (iv) perceptions of, and potential positive and negative impacts to income generating activities such as tourism, farming, agriculture and exports; (v) potential human health benefits due to the reduction of rodents that could vector diseases (Morand, Jittapalapong & Kosoy, 2015); and (vi) the socio-economic and cultural effects of accidental transfer to non-target populations.

5.2.1.2 Potential synthetic biology applications: Management of invasive invertebrates and plants

Priority invasive alien species threats to native species cover the full range of biological taxa (Lowe et al., 2000). Thus, while invasive vertebrates are a large issue, especially on islands, mainland conservation and biodiversity impacts are also frequently incurred from invasive invertebrates and plants, and also introduced disease (Section 5.3.1).

For the management of invasive invertebrates impacting biodiversity, engineered gene drive is arguably closer to realisation than for vertebrates, and likely more applicable due to generally shorter generation times leading to faster spread through populations. Indeed, it has been argued that engineered gene drive for the management of conservation pests should logically be developed first for invasive invertebrates such as common and German wasps (Dearden et al., 2017). Technology development for application to invertebrates is further developed than for vertebrates, both in-silico (mathematical modelling on computers) and in the laboratory. In general, a variety of self-disseminating engineered gene drive systems have been proposed, with many now functional at the proof-of-principle stage in several targeted insect species, predominantly mosquitoes (Sinkins & Gould, 2006; Harvey-Samuel, Ant & Alphey, 2017). It is also generally accepted that there are fewer technological hurdles to overcome (i.e. simpler genetic control of breeding than in vertebrates, making it easier to identify target genes for desired pest control effects), and potentially fewer ethical, social and cultural issues surrounding application for invertebrate as opposed to vertebrate invasive alien species control.

For the management of invasive invertebrates impacting biodiversity, many non-gene-drive synthetic biology control approaches may also be applicable. This is because it is more feasible to breed large numbers of target invertebrates over extended periods of time than target vertebrates. So, approaches such as Release of Insects carrying Dominant Lethals (RIDL) and *Wolbachia* reproductive sex-bias (Appendix 4 (www.iucn.org/synbio)) may have greater potential in this context than for the management of invasive vertebrates.

For the management of invasive plants impacting biodiversity, the synthetic biology approaches currently under consideration for application to vertebrates and invertebrates are less relevant; they only potentially apply to strictly sexually reproducing species, with highest efficiency predicted for short-lived taxa, while many plants are capable of breeding asexually and can be long-lived (e.g. trees). However, researchers are exploring genetic methods for controlling sexually reproducing short-lived weed species such as waterhemp (*Amaranthus rudis* and *A. tuberculatus*) and Palmer amaranth (*A. palmeri*) (Appendix 3 (www.iucn.org/synbio); https://www.eurekalert.org/pub_releases/2018-03/uoic-uii032818.php). Researchers claim that synthetic biology could eradicate invasive plants where conventional approaches cannot, while avoiding non-target impacts of herbicide application. Mechanisms currently being explored are male-biased reproductive sex ratio engineered gene drive approaches similar to those being investigated for invasive animals. For other invasive plants, genetic alterations are being considered to improve the efficacy of traditional biocontrol approaches (Gressel, 2002; Duke, 2003; Tranel & Horvath, 2009).

Case study 2:

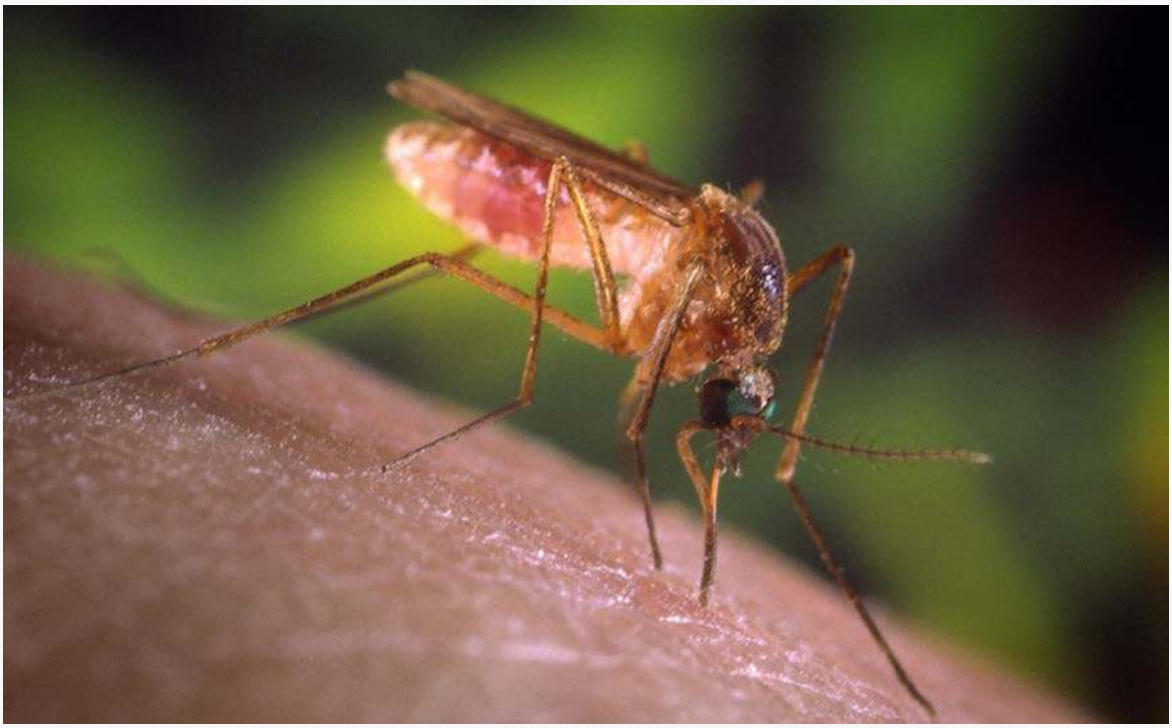
Controlling invasive mosquitoes to prevent bird extinctions in Hawai‘i

Chris Farmer, Brad Keitt

Issue

Native Hawaiian forest birds are among the most threatened in the world. It is widely accepted that introduced mosquito-vectored avian malaria and pox virus are responsible for past extinctions, and ongoing range contractions and declining populations (Atkinson & LaPointe, 2009a; Atkinson & LaPointe, 2009b). No native mosquitoes are present in Hawai‘i. The southern house mosquito (*Culex quinquefasciatus*) was introduced to Hawai‘i in the early 1800s, avian pox arrived in the late 1800s, and avian malaria in the early 1900s. With no prior exposure or natural immunity, the native songbirds were, and remain, highly susceptible to these non-native pathogens transmitted by *C. quinquefasciatus*. These factors contributed to the extinction of more than 24 species of Hawai‘i’s honeycreepers, plus another seven species from other taxa (Pyle & Pyle, 2017) including the extinction of the entire Mohoidae family (Fleischer, James & Olson, 2008).

Presently, almost no native songbirds exist in Hawai‘i at elevations below 1,370 metres, where mosquitoes are abundant. With global warming, mosquitoes are expanding into higher elevation forests and causing rapid declines in many native bird populations (Atkinson et al., 2014; Fortini et al., 2015; Paxton et al., 2016). Mosquitoes are expected to spread to all remaining disease-free forest habitats and cause the extinction of up to 12 species of Hawai‘i’s remaining honeycreepers (Atkinson & LaPointe, 2009a; Atkinson & LaPointe, 2009b; Fortini *et* al., 2015; Paxton et al., 2016) and have a strong negative effect on the remaining native thrushes, flycatchers and corvid (Atkinson & LaPointe, 2009b; Vanderwerf *et al.*, 2006; Atkinson et al., 2014; Fortini et al., 2015). However, populations of three honeycreeper species are showing signs of resistance or tolerance in lowland populations, and might be able to survive an increase in disease prevalence (Woodworth et al., 2005; Krend, 2011; Atkinson et al., 2013).



— **Southern house mosquito (*Culex quinquefasciatus*)** (Gado Images / Alamy.com)

— **‘I’iwi (*Drepanis coccinea*)** (Thomas Chlebecek / Shutterstock.com)

Existing interventions and limitations

Significant effort is dedicated to the conservation of Hawai‘i’s forest birds, including localised predator control, habitat restoration and management, and captive propagation. However, most populations continue to decline (Gorresen et al., 2009; Paxton, Gorresen & Camp, 2013; Paxton et al., 2016; Genz et al., 2018; Judge et al., 2018). For example, two honeycreeper species on Kaua‘i, the ‘Akeke’e (*Loxops caeruleirostris*) and the

‘Akikiki (*Oreomystis bairdi*), both listed by IUCN as Critically Endangered, have shown recent population declines of 89–98 per cent and are projected to become extinct in the near future (Paxton et al., 2016). Maui parrotbill (*Pseudonestor xanthophrys*), or Kiwikiu, is also listed by IUCN as Critically Endangered, with only a few hundred individuals left and is projected to lose 90 per cent of its habitat due to climate change, mosquitoes and avian disease (Fortini et al., 2015; Judge et al., 2018). The ‘I’iwi (*Drepanis coccinea*), likely Hawai‘i’s most iconic bird, is declining and was declared as threatened by the US Fish and Wildlife Service in 2017 and Vulnerable by IUCN (Paxton, Gorresen & Camp, 2013; US FWS, 2017). The species is highly susceptible to avian malaria (Atkinson et al., 1995; Atkinson & LaPointe, 2009a).

Efforts to address the issue of mosquito-vectored avian diseases to protect Hawaiian forest birds are limited, primarily because few tools have been available (LaPointe, Atkinson & Samuel, 2012; Reed et al., 2012). Spray insecticides would cause significant damage to native arthropod populations, and likely have direct negative impacts on forest birds (LaPointe et al., 2009; Reed et al., 2012). Reducing mosquito breeding sites is being attempted, but in what are some of the wettest and most rugged habitats on earth, this is only practical at small scales and is impossible at a landscape scale (LaPointe et al., 2009; LaPointe, Atkinson & Samuel, 2012). It is clear that these conventional mosquito control methods are unlikely to safely and permanently suppress or eradicate mosquitoes and mosquito-borne avian disease in Hawaiian forests (LaPointe et al., 2009; LaPointe, Atkinson & Samuel, 2012; Reed et al., 2012).

Synthetic biology description

The US Fish and Wildlife Service, Hawai‘i Department of Land and Natural Resources, and the American Bird Conservancy are exploring the *Wolbachia* Incompatible Insect Technique (IIT). *Wolbachia* is a naturally occurring genus of bacteria found in 50 per cent of arthropods (Weinert et al., 2015). This technique involves injecting mosquitoes with a novel strain of Wolbachia in a lab and rearing large numbers of infected mosquitoes. While *Wolbachia*-infected males can reproduce with females infected with the same strain of *Wolbachia*, and their offspring will also harbour *Wolbachia*, *Wolbachia* males cannot produce viable offspring with non-*Wolbachia* females or females with a different strain of *Wolbachia* (Atyame et al., 2016). Thus, releasing *Wolbachia* males to mate with wild (non-*Wolbachia*) females can lead to population suppression and even eradication (Zabalou et al., 2004; Atyame et al., 2016; Mains et al., 2016).

Potential adverse effects and limitations

There are two general potential adverse effects to consider. The first is the concern of foreign *Wolbachia* effects on non-target species. However, the IIT has a calculated probability of success and a growing body of evidence that it is safe, based upon extensive trials in other locations with *C. quinquefasciatus* and various *Aedes spp*. (Hoffmann et al., 2011; O’Connor et al., 2012; Atyame et al., 2015, 2016; Mains et al., 2016). Because *Wolbachia* is a naturally occurring endoparasite that is only passed on through sexual reproduction (Atyame et al., 2015, 2016), and only the non-biting males are released, the likelihood of the foreign bacterium being passed to other species is low (Vietnam Eliminate Dengue Project, 2011; US EPA, 2017). The second is the concern that eradication or significant population suppression of native mosquito populations that play important ecological roles could have adverse community and ecosystem effects. This is not a concern in Hawai‘i where all mosquitoes are introduced.

Even though *Wolbachia*-infected males are only fertile with *Wolbachia*-infected females, Wolbachia-infected females are fertile with both infected and uninfected males (Atyame et al., 2015; Mains et al., 2016). Release of a small number of infected females could thus lead to the unintended spread of the *Wolbachia* infection

into the wild population, thereby weakening or preventing the desired population suppression. Therefore, rigorous sex-separation is required before any and all releases (Atyame et al., 2016).

There are significant, substantial and widespread concerns by local stakeholders about the use of synthetic biology to control mosquitoes in Hawai'i. Ultimately, the decision to proceed with field trials will be up to the residents and regulatory agencies of Hawai'i. A key step is a community engagement process that provides opportunities for robust discussion and sharing of information to facilitate informed decisions on the part of the stakeholders.

Socio-economic and cultural considerations

Some potential areas that warrant further study and engagement are: (i) the socio- economic and cultural impact of an increase of tourism from the potential protection of native birds (Department of Business Economic Development and Tourism, 2004); (ii) whether the increased abundance of culturally important birds used for traditional practices would have unintended effects on the islands' social dynamics (Amante-Helweg & Conant, 2009) and (iii) concerns about how synthetic biology approaches could create stress and conflict within local communities and impact their social fabric.

5.2.1.3 Potential adverse effects and limitations

A key influence on the field applicability of synthetic biology approaches for invasive alien species management will be the potential adverse effects of such approaches (Harvey-Samuel, Ant & Alphey, 2017). The critical concern for engineered gene drive application is adverse effects on non-target populations of the same species due to their spread beyond the target population (Marshall & Hay, 2012). In such circumstances, conservation gains achieved through impacts on the targeted invasive population could be offset or even outweighed by conservation losses elsewhere, if populations are impacted where the species targeted is native or performs essential roles in community structure and ecosystem dynamics. Several lines of technical development have been proposed to make engineered gene drive systems self-limiting, such that they can be applied more tactically with reduced potential for spread to non-target populations. Such development is currently only theoretical or at early stages (Appendix 3 (www.iucn.org/synbio)).

There are also concerns for most synthetic biology approaches of adverse effects on non-target species (direct effects on their biology and ecology, and associated community and ecosystem knock-on effects), should there be viable mechanisms for horizontal gene transfer through which new or modified genes could find their way into other species. The reliance on sexual reproduction for such gene transfer (Andersson, 2005) reduces this risk for application to invasive vertebrates, although they need to be front-of-mind for cases where interbreeding between species can occur. When one is considering application to invertebrate and plant invasive alien species, the likelihood of such transfer is higher due to the greater propensity for interbreeding in some species groups and contexts (Moro et al., 2018). However, the potential for gene transfer via interbreeding is generally relatively low, since instances where interbreeding may occur can in most cases be identified from existing knowledge (e.g. Hopper, Britch & Wajnberg, 2006). In addition, multiple genetic changes are generally needed for phenotypic change, and these vary from species to species (Johnson et al., 2016).

Irrespective of the technology employed, should synthetic biology field trials occur, concerns over spread make sites from which organism dispersal is naturally limited, and/or can be effectively limited through management, potentially more appropriate places for testing and initial deployment. Similarly, pest populations with identifiable "private alleles" (alleles found only in the target population) or unique fixed alleles may be more appropriate targets, since drive mechanisms can potentially be self-limited to such population genetic characteristics (Esvelt et al., 2014). Initial trials may also be better conducted in limited

spatial areas (where potential drive mechanisms are less likely to be confronted with scaling-up issues that could lead to the interruption of dissemination through the targeted pest population (Unckless, Clark & Messer, 2016; Champer et al., 2018)) and over limited timescales (to further safeguard both against organism spread away from the target population and the interruption of dissemination). Finally, there should be due consideration of societal and cultural concerns, including impacts on people, their pets, domestic stock, water catchments, animals for hunting and edible plants (Wright, 2011), for any potential trial site.

Since there is as yet no consensus on what type of field sites may be best for trialling genetic biocontrol (James et al., 2018), trial sites need to be considered on a case-by-case basis. Should field trials progress, a lot can be learned from other fields for their design, in particular in health for vaccines and medicines, or for the development of biological control agents. Key technical factors on which such consideration should be based may include characteristics of the target population, the local ecosystem, the characteristics of the modification introduced, the potential for spread, the ability to conduct comprehensive monitoring, and the ability to shut down trials should such a step be required. In addition, conducting formal risk assessment processes prior to any use of synthetic biology for invasive alien species management, be it as a trial or an operational application, would ensure compliance with all relevant laws and regulations, and further ensure that the values of decision makers are explicit in the specific risk assessment. Such risk assessments should include experts from a variety of fields, including conservation scientists and practitioners. Additionally, given the novelty of technologies and approaches being considered, community engagement at all stages of any proposal or project would greatly increase the capacity of all stakeholders, including the general public, to robustly consider the approaches proposed in an informed and open manner. For engineered drive mechanisms, key factors that need to be taken into consideration when assessing their potential viability include the life history, fecundity and generation time of the target invasive alien species and the dispersal and survival ability of introduced animals into extant populations of the target species (Moro et al., 2018).

5.2.2 Reducing pressures from wildlife trade

Unsustainable international commercial trade in wildlife, whether legal or illegal, is one of the greatest threats to wildlife today (Butchart et al., 2010; Nijman, 2010; Duckworth et al., 2012; Challender, Harrop & MacMillan, 2015; Eaton et al., 2015). Wildlife trade affects multiple species, from timber and ornamental plants, to corals, to marine and terrestrial vertebrates. Unsustainable trade, by definition, threatens the survival of the target species, and also the biodiversity of their habitats, since the animals hunted for trade are often keystone species that act as predators, pollinators, dispersers, browsers and ecosystem engineers (Waldram, Bond & Stock, 2008; Blake et al., 2009; Estes et al., 2011; Ripple et al., 2016). Many unsustainably traded terrestrial and marine species are key to local communities, and their loss often threatens the livelihoods of some of the world's poorest and most marginalised people (Cooney et al., 2015). For high value species, international trade is linked to organised crime, and presents security threats to local communities and regions (Wyler & Sheikh, 2013). Illegal trade of such species is facilitated by corruption at all points in the trade chain (Bennett, 2015).

Conventional approaches to addressing unsustainable trade have been numerous. If trade is unsustainable but legal, options are (i) to continue to operate it but put measures into effect to increase sustainability (e.g. quotas, seasonal closures, zoning); (ii) enact legislative change to render the trade illegal and implement programmes of enforcement at all points along the trade chain, from source to market; (iii) reduce demand; and (iv) community engagement and provision of alternative livelihoods. All have had considerable success; however, illegal trade in species with low productivity and high levels of demand, and hence high value, is extremely challenging for management, given the levels of corruption and involvement of organised crime networks.

Increasingly sophisticated, technology-supported systems are being deployed to protect animals at their source (e.g. the enforcement programme SMART; http://smartconservationtools.org/), and to

support intelligence networks around sites and key transportation routes. Where well-resourced, these can be successful at individual sites (WCS, 2018). Demand reduction programmes in key consumer countries are also being undertaken. However, as long as demand for particular wildlife species and products remains high, conserving the target species remains one of the greatest challenges in conservation today.

One approach to supply markets while taking pressure off wild populations is to provide substitutes for wild-caught species. Traditionally, these have come from cultivated (e.g. ornamental plants) or captive bred sources (e.g. tortoises and turtles for the food and pet trades; skins and furs). Under the Convention on International Trade in Endangered Species of Wild Flora and Fauna (CITES), captive-bred specimens from registered facilities can be sold legally, even if wild-caught specimens of the same species cannot be. This can be successful when a verifiable chain of custody prevents laundering of illegally-obtained wild-caught products into legal markets (e.g. peccary skins) (Bodmer, Pezo Lozano & Fang, 2004) or, more commonly, that the cultivated or captive-bred product is preferred over the wild one due to higher quality or lower price (e.g. many ornamental plants, certain reptile skins). This approach is challenging for high value species, especially if their biological productivity is low, given the ease in a corrupt system with which wild-sourced products can be laundered into legal markets (Gratwicke et al., 2008; Bennett, 2015).

5.2.2.1 Potential synthetic biology applications

Synthetic biology has been proposed as another way of producing items demanded in trade while taking pressure off the wild species (Appendix 3 (www.iucn.org/synbio)). If the synthesised item is a perfect substitute for the wild product, this could indeed potentially be highly positive for conservation, taking pressure off the wild species while supplying market demand. A good example is the recombinant Factor C (rFC), a synthetic horseshoe crab blood used by the pharmaceutical industry which replaces the need for the wild product (Case study 8). Squalene is another example; since the oil is used in cosmetics, it is the property of the oil that is important, not its origin. Hence, synthetic substitutes could indeed reduce or remove the need to exploit wild species (Chapter 6.6). In general, the technology for other proposed synthetic biology applications for reducing wildlife trade pressures is still to be developed.

5.2.2.2 Potential adverse effects and limitations

Potential adverse effects of applying such technologies in the context of traded wildlife species arise if the synthesised item is not, in the eyes of consumers, a perfect substitute for the wild-sourced product. For example, for many species in demand for traditional Asian medicines, users frequently believe that wild-sourced products are more efficacious (Gratwicke et al., 2008) and, in the case of bear bile, are willing to pay a premium for a wild-sourced rather than farmed product (Crudge, Nguyen & Cao, 2018). There have been suggestions to manufacture rhino horn using synthetic biology (Africa Geographic, 2015). All trade in rhino horn globally is currently illegal (except for domestic trade within South Africa), with three of the five species of rhino being Critically Endangered and among the most imperilled species on the planet. Opening a legal market for the synthetically manufactured product could prove more harmful than beneficial, because it would render enforcement of illegal trade in wild-sourced horns difficult or impossible, especially when the illegal trade is currently run by corrupt syndicates (Rademeyer, 2012), and when demand is almost inevitably likely to be greater for the wild-sourced product. For further information on potential CITES concerns see the legal analysis by Lyman & Wold (2013), with additional insights into the complexities of wildlife products made by advanced technologies.

5.3 Adaptation

5.3.1 Improving species resilience to threats

Climate change and disease are exacerbating persistent challenges to biodiversity such as habitat destruction, invasive species and overharvesting (Sala et al., 2000). Together these processes can lead to the extensive

fragmentation and isolation of natural populations, with effective population sizes often much smaller than those under historical conditions (Stowell, Pinzone & Martin, 2017). When populations fall to low numbers, they can experience inbreeding depression, whereby the expression of deleterious recessive traits is more likely due to lower gene pool diversity, resulting in reduced fecundity and/or survival. They may also lack the adaptive variation necessary to overcome novel environmental challenges; bottleneck events may even increase the frequency of deleterious genetic variants in the remaining population (Marsden et al., 2016).

Bottleneck populations may experience an increase in susceptibility to disease (Hale & Briskie, 2007; Tompkins, 2007) or an increased frequency of genetic disorders that negatively impact survival (Räikkönen et al., 2009). Moreover, because many small populations are isolated by physical, environmental and ecological barriers, they may have few or no opportunities for infusions of novel genetic variation via gene flow. Habitat protection could help to increase population size, but for populations trapped in an extinction vortex (Gilpin & Soulé, 1986), habitat protection alone may be inadequate for successful conservation (Stowell, Pinzone & Martin, 2017). Where the proximate threat to inbred populations is disease, conventional vaccination/treatment approaches can protect small numbers, but are increasingly unrealistic as the spatial scale over which populations occur increases (Cross, Buddle & Aldwell, 2007). In this context, fungal pathogens such as chytrid fungus threatening amphibians globally (Fisher, Garner & Walker, 2009; see Box 5.1) and white-nose syndrome in North American bat species (Blehert et al., 2009) are proving particularly intractable to conventional management approaches (Fisher et al., 2012).

Box 5.1
Future challenge: The potential use of synthetic biology to control lethal fungal pathogens of amphibians

Reid Harris & Louise Rollins-Smith

Note that this is a Future Challenge and not a Case study, as there is no current synthetic biology solution under development for this application.

Issue

Increasingly, there are major challenges to biodiversity conservation with no obvious solutions realisable in the time-scale necessary to make the difference needed. Examples include emerging infectious diseases and the impacts of climate change and ocean acidification. Researchers are searching for new technologies that might overcome these challenges; synthetic biology approaches have promise, but for most proposed applications we currently lack clear evidence for their usefulness and safety. We present some of the considerations for the design of a research agenda to explore the potential of synthetic biology approaches as solutions to one such challenge.

Chytridiomycosis is a fungal disease of amphibian skin that evolved in eastern Asia and emerged elsewhere in the early 20th century, which coincided with the global expansion of commercial trade in amphibians (O'Hanlon et al., 2018). This disease has led to widespread mortality and extinction; for example, approximately 41 per cent of amphibian species in a montane region of Panama declined or went extinct once the causative agent, *Batrachochytrium dendrobatidis* (*Bd*), arrived (Crawford, Lips & Bermingham, 2010). It is thus considered the greatest disease threat to biodiversity (Wake & Vredenburg, 2008). A second chytrid fungus, *B. salamandrivorans* (*Bsal*), has caused population extinctions of the fire salamander *Salamandra salamandra* in Europe (Martel et al., 2013; Stegen et al., 2017). While currently not found in North America, susceptibility trials of native North American salamanders reveal that some species are lethally affected, including all tested species in the newt family (Martel *et al.*, 2014). Given this, with North America being home to the largest number of salamander species globally, accidental introduction of *Bsal* could drastically reduce amphibian biodiversity and result in concomitant ecosystem effects.

Amphibians are major parts of ecological communities worldwide (Hairston & Hairston, 1987); for example, the biomass of salamanders in one North American forest was estimated to be 2.5 times that of all breeding birds and equal to that of small mammals (Burton & Likens, 1975). This estimate was based on surface counts and

as such is an underestimate of the salamander biomass relative to birds and mammals. Removal experiments have shown that decimation of terrestrial salamanders would lead to CO_2 release due to accelerated leaf decomposition caused by the release of leaf-shredding invertebrates from predation (Best & Hartwell, 2014; Hickerson, Anthony & Walton, 2017), potentially contributing to global warming (Wyman, 1998). Salamanders are also keystone species in temporary ponds; some frog species are greatly reduced in abundance if salamanders are removed (Morin, 1983).

— **Fire salamander (*Salamandra salamandra*)** (Beatrice Prezzemoli / Shutterstock.com)

— **Barking tree frog (*Hyla gratiosa*)** (Jay Ondreicka / Shutterstock.com)

Existing interventions and limitations

Two proposed non-synthetic-biology strategies to protect amphibians from chytridiomycosis are 'vaccination' with *Bd* or *Bsal* antigens (or attenuated strains), and the augmentation of naturally-occurring anti-chytrid skin bacteria (Bletz et al., 2013; McKenzie et al., 2018). A vaccination strategy could involve infection and 'cure' with heat or antifungal treatment (McMahon et al., 2014). Bioaugmentation has had success in laboratory trials and in one field trial, and new 'omics' analysis technologies (i.e. metagenomics, transcriptomics and metabolomics) might lead to better selection of probiotic strains for clinical trials (Rebollar et al., 2016). Both strategies may require considerable resources to bring to large-scale efficacy.

Potential synthetic biology solutions

Several synthetic biology approaches to counteract *Bd* and *Bsal* are being considered. First, can the pathogens be genetically modified to become avirulent? This might be possible using CRISPR-Cas9 gene editing, but it is not yet known what critical virulence factors are responsible for amphibian deaths, or whether avirulent strains of *Bd* or *Bsal* could displace or protect against virulent strains. Virulence appears to derive from growth rate (Mitchell et al., 2008; Fisher, Garner & Walker, 2009), so it is likely that virulent strains would be more competitive. Also, *Bd* and *Bsal* are asexual; thus, a gene drive mechanism would not be successful in spreading modified genes beyond one clonal line. Hybridisation has been inferred by genetic analyses of *Bd* strains; hence sexual reproduction must have occurred in the past but it has never been observed (Schloegel et al., 2012; Greenspan et al., 2018).

Second, might gene editing techniques be used to modify the host species? Three components of amphibian defences can be considered: innate immunity, acquired immunity and the microbiome. There is evidence in some species that antimicrobial peptides (AMPs) of the innate immune system that are protective against *Bd* are secreted into the epidermal mucus (Woodhams et al., 2007). It might thus be possible to edit AMP genes into the genomes of species that lack them. For example, many ranid frog species have well-developed AMP genes, but bufonids (toads) appear to lack them (Conlon, 2011); if they were to be introduced into bufonid species, promoters would be needed that assure their expression in skin glands. CRISPR-Cas9 methods for deletion of genes and "knock in" of genes have been developed for the frog species *Xenopus laevis*, and could theoretically be developed for other amphibians (Banach et al., 2017).

The amphibian acquired immune system is as complex as in all other higher vertebrates, and thus it would be difficult to identify a limited set of components to modify by gene-editing to provide protection against chytridiomycosis (Flajnik, 2018). There are also significant problems with the inducement of breeding in non-model species, to supply eggs and sperm for genome manipulation (Trudeau et al., 2013; Clulow et al., 2014).

With regards to the microbiome, there is strong evidence in some species that skin bacteria secrete metabolites that can protect against *Bd* (Harris et al., 2009). However, probiotic addition experiments have had mixed success largely because the probiotic species do not persist (Becker et al., 2011). The genetic basis for production of some of these protective metabolites is known (August et al., 2000). Thus, the persistence issue could potentially be addressed by inserting such genes into skin bacteria that naturally occur at high abundance. For this approach to be successful the genetically modified bacteria would have to displace the unmodified members of the same species, the likelihood of which is unknown.

Synthetic biology approaches may thus help prevent continued losses of amphibian species due to chytridiomycosis. A next step would be to evaluate and compare approaches that do and do not involve synthetic biology, incorporating many of the considerations outlined above.

Another strategy that aids the conservation of populations facing extinction is the deliberate introduction of individuals as vehicles for the infusion of novel alleles. This strategy has been variously termed genetic rescue, facilitated migration, intentional hybridisation or introgression, and admixture rescue, and is the topic of much discussion (Tallmon, Luikart & Waples, 2004; Whiteley et al., 2015). For simplicity, this chapter refers to actions taken by conservationists to increase gene flow, genetic diversity and fitness as "genetic rescue." A suite of studies highlight the value of genetic rescue in increasing population fitness (Frankham, 2015), demonstrating the power of innovative methods for saving struggling populations. However, such approaches are limited to using the genetic variation remaining in extant populations; for many severely bottlenecked species this will likely be insufficient to prevent ongoing population decline.

In recent years it has been proposed that genome editing, most recently using the CRISPR-Cas9 toolkit, might be applied to address this issue, for

example by attempting to alter or introduce genes with the goal of enhancing species survival against specific threats, including disease and climate change (Appendix 3 (www.iucn.org/synbio)). Several such applications have been proposed, as detailed below. Other synthetic biology approaches are also being pursued, such as improving general species viability through reintroducing extinct genetic variation stored in 'frozen arks' (and potentially also museum specimens) back into extant populations (cloning; Appendix 4 (www.iucn.org/synbio)). Note that it is conceivable that synthetic biology could also contribute to the restoration of ecosystem resilience and function, e.g. through the resilience or restoration of keystone species, and thus the prevention of ecosystem collapse (Bland et al., 2015).

5.3.1.1 Potential synthetic biology applications: Improving general species viability

Cloning approaches are being attempted, for example, to increase white rhino genetic diversity (Hildebrandt et al., 2018). The northern white rhino (*Ceratotherium simum cottoni*) population is down to just two infertile females; the last male died in March 2018. The southern subspecies is some 21,000 animals strong. Yet the genomes of northern animals, albeit based on a handful of samples, are more diverse. Researchers are attempting to create embryos by injecting northern white rhino sperm nuclei from frozen material into southern white rhino unfertilised eggs; to date they have survived to only an early embryonic developmental stage (Hildebrandt et al., 2018). Similar approaches are being considered for other endangered species including the black footed ferret (Case study 3), yet in most cases such approaches are currently only speculative.

5.3.1.2 Potential synthetic biology applications: Improving species resilience against disease

Using synthetic biology approaches to improve species resilience against disease has been proposed for several species such as the black-footed ferret threatened by sylvatic plague (Case study 3), both Asian and African elephants threatened by elephant endotheliotropic herpes virus, amphibians and salamanders globally threatened by chytrid fungus,

— Northern white rhinoceros (*Ceratotherium simum cottoni*) (EcoPrint / Shutterstock.com)

and bats in North America threatened by white-nose syndrome (Redford, Adams & Mace, 2013).

Most proposed applications are currently speculative (Appendix 3 (www.iucn.org/synbio)), but a clear demonstration of potential has been made for resilience against a tree pathogen (Case study 4). The American chestnut was nearly wiped out by chestnut blight; research indicates that a synthetic biology solution is technically ready for field testing (Steiner et al., 2017). Because the chestnut is fast-growing, long-lived and resistant to decay, restoration of American chestnut using blight resistant stock could potentially increase carbon sequestration or storage in forested landscapes (Gustafson et al., 2017). However, carbon dynamics are also affected by interspecific competition, succession, natural disturbance and forest management activities, and it is unknown how chestnut restoration might interact with these processes (Schmidt et al., 2011).

Case study 3:
Synthetic biology to address conservation threats to black-footed ferrets

Tom Maloney, Ben Novak

Issue

The black-footed ferret (*Mustela nigripes*) was once an ecologically important and widespread small carnivore in the Great Plains of North America. Efforts throughout the 20th century to eradicate the ferret's chief food source, the prairie dog (*Cynomys spp.*), resulted in dramatic declines to near extinction by the late 1970s (Biggins and Schroeder, 1988). Twice presumed extinct, a population of ferrets in Wyoming was re-discovered in the early 1980s. The US Fish and Wildlife Service (US FWS) began a recovery effort in 1985. Traditional conservation approaches, including habitat protection and careful pedigree management in captive breeding including artificial insemination (Howard et al., 2016), have helped the recovery of the species. However, the species remains threatened by extinction.

Existing interventions and limitations

Recovery efforts have enabled the reintroduction of hundreds of ferrets within the former range, but all are descended from a founding population of just seven individuals. Two principal threats are seen as critical to address in order to achieve the sustained recovery of the species: genetic drift/inbreeding depression and susceptibility to sylvatic plague, a widespread non-native disease (Antolin et al., 2002; Abbott et al., 2012). Genetic drift has resulted in a loss of 15 per cent of original founding genetic diversity in the current ferret generation (Wisely et al., 2015).

Black-footed ferrets have no innate immunity to sylvatic plague, which causes high mortalities in wild populations (Roelle, 2006; Matchett et al., 2010). The US Geological Survey developed a vaccine for sylvatic plague, which US FWS applies in a labour-intensive programme to vaccinate released ferrets (Abbott et al., 2012). However, it is difficult to deliver this vaccine (which requires a booster) to wild-born ferrets. Hence, at least at this stage, continued vaccination of both captive and wild-born ferrets is not a path to sustained recovery.

Synthetic biology description

Scientists at the San Diego Frozen Zoo had cryopreserved two cell lines from the last wild ferrets in Wyoming. Whole genome sequencing conducted in 2016 revealed that those cell

Black-footed ferret (*Mustela nigripes*) (Kerry Hargrove / Shutterstock.com)

lines have significant genetic variation that could be used to restore lost genetic diversity (Revive & Restore, San Diego Zoo Global and Intrexon, 2016). Incorporating this variation would effectively introduce two new founders to the extant population (Wisely et al., 2002). In July 2018, the US Fish & Wildlife Service issued an endangered species recovery permit to authorise the development of laboratory methods to clone the frozen cell lines.

There is also now the potential to use precise genome editing techniques to create inheritable immunity to plague (Novak, Maloney & Phelan, 2018). Research has shown that plague immunity is antibody mediated (Hill et al., 2003; Liu et al., 2017) and that black-footed ferrets have plague antibodies. The data from all plague challenges and exposures of non-vaccinated ferrets show that the antibodies respond slowly during an infection; only exposure to the vaccine brings about antibody expression at the early stage of infection (for subsequent exposure). It may now be possible to duplicate the plague-specific antibody genes that are triggered by vaccination in a manner that would produce lifelong expression of plague-antibodies, a process known as vectored immunoprophylaxis (Sanders & Ponzio, 2017). The above referenced permit also authorised efforts to test the viability and efficacy of genome editing to activate innate alleles to upregulate the antibody response and convey inheritable resistance to sylvatic plague in black-footed ferrets. A second, transgenic approach might be to edit plague-resistant alleles from the domestic ferret (for which plague is not fatal) into the genome of the black-footed ferret (Novak, Maloney & Phelan, 2018).

Testing of these approaches is planned to be conducted first in laboratory mice for efficiency, since they have short generation times. If successful, methods and fitness testing could then be expanded to testing in black-footed ferrets. An experimental population of genome-edited, disease-resistant black-footed ferrets could then be established to assess responses to plague and verify immunity. The fitness of this experimental group of ferrets would need to be carefully analysed over several generations, specifically replicating environmental conditions that black-footed ferrets face in the wild, to

confirm the safety of integrating plague-immunity throughout the entireferret population.

Potential adverse effects and limitations

While these novel tools hold great promise, there are still many uncertainties. The use of the very-closely-related domestic ferret as a surrogate parent for the cloned cell lines is untested. There may be incompatibility issues between the species (Wisely et al., 2015). The development and field-testing of vectored immunity techniques in ferrets is also uncharted territory and will take years to implement because long-term fitness testing is required to rule out any unexpected effects. However, these considerations are required steps to realise the potential of recent developments in synthetic biology to address widespread challenges that make the black-footed ferret conservation dependent.

Socio-economic and cultural considerations

This species was one of the first endangered mammals to be listed in the US, and the public has a strong interest in its successful recovery. Regulators are embracing a deliberate and purposeful public engagement process to provide every opportunity for concerned stakeholders to participate in proposed recovery efforts. The potential recovery of the black-footed ferret could have a significant economic impact through recovery of the grassland/prairie ecosystem. Further investigations are required to analyse other indirect socio-economic impacts of this synthetic biology application, including the potential socio-economic impact of replacing existing conservation approaches. There may also be ethical objections to modifying populations of endangered species, although the proposed effort has prioritised the enhancement of innate alleles over a transgenic approach, since these are already part of the black-footed ferret genome. Ranchers in the US have expressed their interest in the recovery of this species because of the land-use restrictions related to the protected status of the ferret (https://www.denverpost.com/2013/02/17/ranchers-sought-to-help-black-footed-ferret/).

Case study 4:

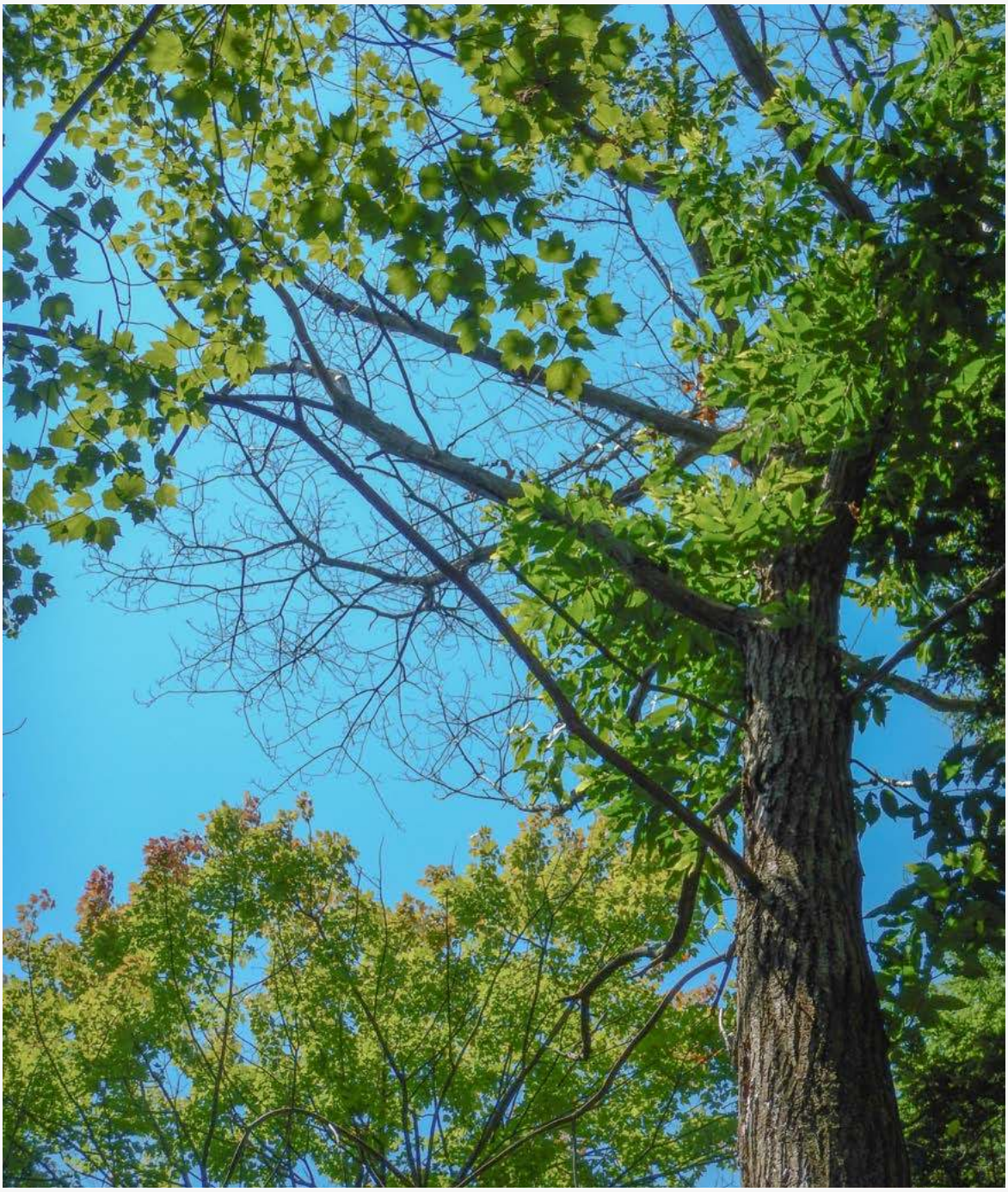
Transgenic American chestnut for potential forest restoration

Andrew E. Newhouse, William A. Powell

Issue

American chestnuts (*Castanea dentata*) once provided sustenance and shelter for wildlife and a healthy and profitable nut crop for humans in the US (Jacobs, Dalgleish & Nelson, 2013). These trees were large and long-lived compared to other species in America's eastern forests (Woods & Shanks, 1959), but were almost entirely wiped out when an invasive blight fungus was accidentally introduced to the United States in the late 1800s (Anagnostakis, 1987).

American chestnuts are not extinct; however, they are categorised as Critically Endangered on the IUCN Red List (http://oldredlist.iucnredlist.org/details/62004455/0) and are generally limited to surviving as small seedlings or stump sprouts, rarely reaching maturity before blight



American chestnut tree (*Castanea dentata*) (William Ragosta / Alamy.com)

reinfection occurs (Paillet, 2002). The American chestnut will not survive indefinitely: without human intervention, wild populations of pure American chestnut will likely continue to decline until they are all but gone (Paillet, 1993). Relatives of the American chestnut in Asia that evolved with the blight fungus are usually able to tolerate blight infections without much damage (Jaynes, 1964).

Existing interventions and limitations

Multiple efforts have been made to breed American with Chinese chestnuts to obtain desirable characteristics from both species, but traditional breeding is a slow and unpredictable process, limited by undesirable traits from the non-native Chinese chestnut (Woodcock et al., 2017). The American Chestnut Foundation backcross breeding program (www.acf.org) shows promise in producing trees with American chestnut growth traits,

but since blight resistance in Chinese chestnuts is controlled by several genes, inheritance by future generations of chestnuts is inconsistent (Steiner et al., 2017). Recent technological advances in genomic screening are improving this process, but it will likely require multiple generations of breeding (Steiner et al., 2017), and blight resistance in backcrossed offspring will logically never surpass that of the Chinese chestnut ancestor (Woodcock et al., 2017).

Synthetic biology description

Researchers at the College of Environmental Science and Forestry in Syracuse, New York, have produced American chestnut trees that show promise to tolerate blight infections (Zhang et al., 2013). This was achieved by inserting a single gene from wheat into a new line of American chestnut trees (Zhang et al., 2013). The same gene, which is found in many other wild and domestic plants, produces an enzyme — oxalate oxidase —that does not kill the fungus but rather breaks down the toxin oxalic acid, which is produced by the fungus and kills American chestnut tissues (Chen et al., 2010). Since this enzyme has no direct fungicidal properties, selective pressure is reduced or eliminated. This effectively means that all potential plantings of transgenic chestnut trees would act as refugia, so the blight fungus is much less likely to evolve resistance over time (NRC, 1986).

Potential adverse effects and limitations

Frequent concerns regarding introducing a transgenic tree into the environment include the genetic diversity of the restored population of American chestnuts and environmental safety to surrounding organisms. Outcrossing lab-produced transgenic trees with surviving wild American chestnuts has the potential to incorporate the necessary genetic diversity and regional adaptations in future generations of American chestnuts, while also protecting them from chestnut blight (Steiner et al., 2017; Westbrook, 2018). Transgenic chestnuts have been tested for safety to many other organisms, including ectomycorrhizal fungi (symbiotic fungi associated with roots that aid in water and nutrient uptake), tadpoles which consume leaf litter, and native seeds, and tests to date have shown no adverse effects compared to traditional breeding (D'Amico et al., 2015; Goldspiel

— **Chestnusts in hand** (nocostock / Shutterstock.com)

et al., 2018; Newhouse et al., 2018). Nutrition testing on the nuts that people and animals would consume has confirmed that nutrition is equivalent to non-transgenic chestnuts. This testing has been completed but is awaiting publication.

Before any restoration with transgenic trees could take place in the US, regulatory approval must be received from at least three different federal agencies: Department of Agriculture, Environmental Protection Agency, and Food Drug and Administration. Specific mandates vary by agency, but the overarching goal is to ensure the transgenic product is not significantly riskier than similar products produced with traditional breeding. In addition to the comparatively well-defined regulatory requirements, there are also unique considerations regarding the public's acceptance of a genetically engineered product intended for release in the wild. Compared to concerns surrounding genetically engineered food crops in commercial agriculture, the non-profit, non-agricultural goals of restoration suggest reduced public concerns about corporate motives. But the long-term reality of introducing to the wild a forest-type tree might warrant different risk analyses than those required by an annually-harvested food crop.

There is widespread potential for related synthetic biology tools to rescue other threatened wild species, including forest trees such as ash (Palla & Pijut, 2015; Lee & Pijut, 2017) and elm (Newhouse et al., 2007). Biotechnology certainly is not the only tool available to protect trees from environmental threats, but the case of the American chestnut indicates that it can potentially be a means to restore healthy and resilient trees to native ecosystems.

Socio-economic and cultural considerations

Research on socio-economic and cultural considerations should be carried out to identify the specific benefits or adverse effects of the restoration of the American chestnut. Some economic considerations around the potential revitalisation of the chestnut value chain could warrant further research, considering the potential adverse effect on this value chain if consumers reject what would be considered as a genetically modified product. The social and cultural impacts of this new approach on existing grassroots movements such as the American Chestnut Foundation for the restoration of this tree would require further assessment and engagement. However, the foundation's leadership and membership are increasingly considering synthetic biology to be a valid and promising means of potential restoration (Steiner et al., 2017).

A small number of large-scale empirical surveys have been conducted on public opinion regarding the use of biotechnology for tree restoration or forest health (Hajjar & Kozak, 2015; Kazana et al., 2015; Needham, Howe & Petit, 2015; Fuller et al., 2016; Jepson & Arakelyan, 2017). These surveys took place in the US, UK and Canada, and a general consensus emerges: in the face of a concrete, human-caused threat, like chestnut blight, public acceptance of biotechnology solutions is generally similar to acceptance of traditional breeding or planting of non-native species, and often more acceptable than taking no action. This reinforces general responses frequently received by College of Environmental Science and Forestry chestnut team members: a small minority of people are sceptical about breeding with Asian chestnuts, another minority is sceptical about genetic engineering, but most people support the idea of restoring American chestnuts by whatever means are safe and effective.

5.3.1.3 Potential synthetic biology applications: Increased resilience to climate change

It has been proposed that synthetic biology solutions could help enable species survival in the face of otherwise intractable threats such as climate change. In cases where species are unable to naturally adapt in a sufficient time-frame for survival, or disperse in either natural or assisted fashion (Ewen, 2012) into areas suitable for survival as native ranges become unsuitable through climate (or indeed other

environmental) change, synthetic biology approaches may be able to confer sufficient resilience to allow ongoing viability within the native range. Such adaptation has been the subject of much research for agricultural plants (Hunter, 2016), for example the creation of drought-tolerant maize (Marshall, 2014).

For conservation application, an example is seen in the fight against mass bleaching of coral reefs as a result of ocean warming (Case study 5). With synthetic biology, the alleles that provide resilience to ocean warming in certain species of coral could potentially be assimilated into the genomes of non-resilient species, reversing the loss of coral reefs around the world on a larger scale (van Oppen et al., 2015; Levin et al., 2017; Cleves et al., 2018). While considerable technological development is still required before synthetic biology can be applied to corals and their microbial symbionts, early achievements suggest such manipulations are possible.

Case study 5:
Corals and adaptation to climate change/acidification

Madeleine van Oppen

Issue

Coral reefs around the globe are being lost at an alarming rate due to a number of factors including climate change, declining water quality, crown-of-thorns starfish outbreaks, coastal development and overharvesting. Climate change is believed to be the biggest threat to the persistence of coral reefs, particularly since the heat waves of 2014–2017 assaulted coral reefs worldwide, resulting in the third global mass bleaching event and extensive coral mortality. For instance, approximately 50 per cent of the coral was lost from Australia's Great Barrier Reef in just two years (2016 and 2017) when the reef experienced extreme summer temperatures (Hughes et al., 2018). Further warming will almost certainly occur within this century, with models showing only a 5 per cent chance that the global temperature increase since pre-industrial times will be less than 2°C by 2050 (Raftery et al., 2017). Thus, ensuring coral reef persistence into the future until global warming is curbed might require alternative interventions that either reduce bleaching stress (such as cooling reef water or shading the reef) or increase coral bleaching tolerance (i.e. bio-engineering solutions).[1]

Existing interventions and limits

Elevated temperatures are known to cause oxidative stress in the coral host animal and its associated microalgal symbionts, triggering a cellular cascade and culminating in the loss of the algae (*Symbiodiniaceae* spp.) from the coral tissues (i.e. coral bleaching) (Weis, 2008). A number of traditional manipulations are being explored to increase coral climate resilience, including selective breeding, interspecific hybridisation, assisted gene flow and probiotics (van Oppen et al., 2015, 2017). Preliminary results are promising (Dixon et al., 2015; Chakravarti, Beltran & van Oppen, 2017; Damjanovic et al., 2017; Chan et al., 2018), but it is not yet clear whether these interventions can achieve the required results in time and at an appropriate scale. Therefore, researchers are assessing and developing

[3] The following paper was published too late to be included in this assessment but should be consulted: National Academies of Sciences, Engineering, and Medicine. 2018. A Research Review of Interventions to Increase the Persistence and Resilience of Coral Reefs. Washington, DC: The National Academies Press. doi: https://doi.org/10.17226/25279

Coral reef bleaching (Sabangvideo / Shutterstock.com)

genetic engineering and synthetic biology options in parallel with traditional approaches.

Synthetic biology considerations

Genetic engineering and gene editing tools may be used to insert coral or microbial genes encoding antioxidant enzymes (Levin et al., 2017) or to introduce gene pathways or synthetic microbes able to produce non-enzymatic antioxidants (see Appendix 3 (www.iucn.org/synbio)). Other genes involved in the thermal stress response may also prove to be useful genetic engineering targets to enhance thermal tolerance (van Oppen et al., 2017). Alternative synthetic biology approaches may be developed to prevent algal symbionts from becoming parasitic during heat stress (Baker et al., 2018).

Crown-of-thorns starfish (*Acanthaster* spp.) are among the largest predators of scleractinian (stony) corals in the Indo-Pacific and adult animals can kill whole coral colonies (Pratchett et al., 2014). High densities of the starfish cause rapid loss of corals and decline of coral reefs (Kayal et al., 2012); for instance, they were responsible for the loss of about 21 per cent of the approximately 51 per cent of coral lost from the Great Barrier Reef between 1985 and 2012 (De'ath et al., 2012). The starfish are highly fecund and can therefore spread rapidly among reefs, sometimes leading to devastating population outbreaks (Babcock & Mundy, 1992). Current biocontrol methods for the starfish involving lethal injection of adults, hand-picking and barriers are only effective over small spatial scales (Hall et al., 2017). The recent identification of crown-of-thorns-specific peptides used in intra-specific communication may lead to the development of starfish traps with a larger-scale impact (Hall et al., 2017). CRISPR-Cas9 mediated engineered gene drive systems with the aim to reduce population growth through, for instance, reduced reproductive rates may also be developed as a biocontrol mechanism effective over large spatial scale.

Potential adverse effects and limitations

Genetic engineering methods are poorly developed for corals and their microbial symbionts. The recent development of genomic resources for *Symbiodinium* and the CRISPR-Cas9 gene editing technology, however, provide promising new avenues for genetic engineering of these dinoflagellates (Levin et al., 2017). For bacteria, well-established systems exist for knocking out, altering and introducing genes in taxa closely

— **Crown-of-thorns starfish (*Acanthaster* spp.)** (Richard Whitcombe / Shutterstock.com)

related to those known to associate with corals (Blackall, Wilson & van Oppen, 2015), including *Vibrio* (Dalia et al., 2017), *Serratia* (Ito et al., 2017) and *Rhodobacter* (Swainsbury et al., 2017). A huge step forward was recently achieved in gene editing by the successful introduction of mutations targeted to three genes of the coral *Acropora millepora* by injecting zygotes with CRISPR-Cas9 ribonucleoprotein complexes (Cleves et al., 2018). While considerable technological development is still required before genetic engineering methods can be applied to corals and their microbial symbionts, these early achievements suggest such manipulations are within the realm of possibility.

As with many proposed synthetic biology applications, there are potential non-target population and species concerns (effects on their biology and ecology, and associated community and ecosystem knock-on effects). Non-target population effects could arise should genetically modified stages disperse from the populations targeted for management to other populations of the same coral host or symbiont species. Non-target species effects could arise, should there be viable mechanisms for horizontal gene transfer, through which new or modified genes (and potentially their effects) could find their way into other species.

For any application of gene drive systems to suppress crown-of-thorns starfish population growth, additional potential concerns that need to be addressed include non-target population effects should modified individuals or their offspring spread from target populations, inadvertent population extinction rather than any desired suppression effect, and any wider ecological implications of both eventualities.

Socio-economic and cultural considerations

A specific socio-economic and cultural considerations assessment would be required for considering a particular intervention in a given ecosystem and context. Some of the socio-economic considerations around the impact of reducing coral bleaching could be particularly relevant to local fisheries (Kittinger

et al., 2015) as well as tourism activities (Spalding et al., 2017). The potential impacts on coastal protection – economic as well as cultural and social for communities traditionally living in these areas – should be further evaluated (Creel, 2003).

5.3.1.4 Potential adverse effects and limitations

Any approach to enhancing genetic variability in a species ("genetic rescue") can be controversial because it is hard to predict how a population will be affected by a migration event (Stowell, Pinzone & Martin, 2017). In some cases, genetic rescue has lowered the fitness of a population by swamping the population or increasing rare deleterious alleles (Hedrick & Garcia-Dorado, 2016); in others, rescue may only be a short-term solution. Some reviews clearly show that genetic rescue has worked (Frankham, 2015); others argue that genetic rescue could create unforeseen problems for the target species and that it overlooks the underlying problems that push species to the brink of extinction (Poppick, 2018). There are also perceived moral barriers to genetic rescue, with some members of the public expressing concern for the taxonomic integrity or "naturalness" of species (Stowell, Pinzone & Martin, 2017), and concern that such interventions are a "slippery slope". For example, if scientists insert one gene, why not more? When does it stop? This is especially problematic when considering endangered species.

There are no immediately identifiable potential adverse effects associated with utilising cloning to improve species resilience, although there are three key limitations (IUCN SSC, 2016): (i) it is currently a relatively inefficient process for many species; (ii) clones in some species have had a tendency towards developmental abnormalities and premature aging, leading to suffering and to short lives, which has led to significant ethical concerns that would offset potential benefits; and (iii) cloning is fully dependent on the availability of intact somatic cells that have been stored appropriately or used directly from living individuals.

CRISPR-Cas9 genome editing has accelerated and facilitated synthetic biology. Yet, it should be acknowledged that targeting just a few genes or genomic regions for editing may not always be sufficient for a phenotypic change, or at least in the way intended for conservation (Johnson et al., 2016). An increasing number of studies have shown that the genetic architecture of many fitness-related traits is largely under the control of many genes of small effect, or polygenic, including the influence of genetic epistatic interactions and functional intergenic regions (Taylor & Ehrenreich, 2015). Therefore, although significant changes in phenotype have been produced with genome editing and transgenesis, including complex phenotypes such as behaviour, there are still significant challenges. Yet, new genomic technologies including CRISPR-Cas9 have great promise for also making it much easier to link genotypes with phenotypes and fitness in non-model species (Bono, Olesnicky & Matzkin, 2015). A clear demonstration of increasing resilience to plant disease has been made, but all animal work is currently speculative or at the early stages of progress.

Existing research identifies a variety of potential adverse effects that should be examined, including for example adverse effects involving non-target impacts and gene flow (Vettori et al., 2016). Where synthetic biology is used to alter the fundamental niche of a species (the entire set of conditions under which it can survive and reproduce itself), that it could potentially alter the ecological and evolutionary trajectories for that species (with potentially deleterious long-term consequences; e.g. a climate change adaptation is engineered, and climate change is eventually reversed) should also be considered.

5.3.2 Creating proxies of extinct species

There have been five mass extinction events in the past 600 million years. In the worst, 250 million years ago, 96 per cent of the marine species and 70 per cent of the land species died off. It took millions of years to recover (Benton, 2015). Many scientists are now predicting we are placed for a sixth mass extinction (e.g. Ceballos,

Ehrlich & Dirzo, 2017; Ceballos & Ehrlich, 2018). It has been estimated that currently three species on the planet go extinct every hour, and that this rate is orders of magnitude higher than the planet has seen in previous catastrophic extinction events (Kolbert, 2014). In this light, solutions are being explored to prevent extinction of endangered species, or even resurrect extinct species, especially keystone species that play a unique and crucial role in the way ecosystems function.

De-extinction, or species revival, is the development of functional proxies for species which previously went extinct (IUCN SSC, 2016). Traditional methods to restore species involve: (i) selective or back breeding: this is essentially a version of existing domestic animal breeding. It is slow, and while it can result in an organism that looks like the extinct species, its genetic code, hence also the ecological functions it performs in the ecosystem, may be quite distinct; (ii) animal cloning: this requires the transfer of the nucleus of the adult cell of an extinct species (e.g. frozen tissue) into the unfertilised egg of a host animal cell from which the nucleus has been removed, creating a true clone (identical to the parent). However, the rate at which DNA degrades makes cloning possible only for relatively recently extinct animals, for which a suitable closely-related host is available. IUCN has developed guidelines for these more conventional techniques (IUCN SSC, 2016).

It has been proposed that species' functional proxies might support ecosystem recovery by restoring ecological function, restarting latent ecological processes and restoring lost ecosystems or ecosystem states (Estes, Burdin & Doak, 2016). In turn, this might increase ecosystem stability in the face of environmental change, promoting network diversity and reducing loss of other species (IUCN SSC, 2016).

5.3.2.1 Potential synthetic biology applications

The technological approaches for the creation of proxy species are currently speculative or at the early stages of development (Appendix 3 (www.iucn.org/synbio)). However, de-extinction has caught the public imagination through high-profile publications and events (TEDxDeExtinction, 2013), high profile projects such as the passenger pigeon project (Revive & Restore, 2018) and media fascination with bringing back woolly mammoths, the ground sloth and other extinct species.

Applying synthetic biology techniques for de-extinction is also hugely complex, and the technical challenges of fully and accurately sequencing the genomes of extinct species are immense (Shapiro, 2015). Although whole-genome sequencing technologies have become more accessible, allowing for the generation of genomic datasets for multiple individuals in species of conservation concern, additional advancements are needed in order to decipher the genomic architecture of complex traits important for species persistence (Johnson et al., 2016).

5.3.2.2 Potential adverse effects and limitations

Creating proxies of extinct species could possibly distract attention and funding from more pressing issues and cost-effective conservation actions to conserve extant but threatened species and ecosystems (IUCN SSC, 2016). The financial and human resource costs of creating a proxy species, introducing it to the wild, and monitoring its progress, would likely be considerable, and could divert resources from the conservation of extant species. It pits an optimistic world of high-tech 'precision' conservation against a more conventional vision of biodiversity conservation achieved primarily through protected areas (Adams, 2017), and broader species and landscape management and planning.

Another proposed adverse effect of resurrecting an extinct species is that it might diminish extinction itself, posing a moral hazard by changing public perceptions in a way that could undermine current and future conservation efforts (IUCN SSC, 2016). The creation of an apparent techno-fix to the crisis of species extinctions and biodiversity loss could have the perverse effect of making society feel better about its throwaway attitude towards nature (DeSalle & Amato, 2017). The social and health welfare of individual animals during the process is a further concern. The severe welfare concerns in relation to processes around the production of animal clones are well

documented. Concerns include the potential suffering of new individuals and of gestational surrogates, the provision of appropriate animal husbandry, social and psychological environments for the species, and the post-release survival of animals following translocation into a novel ecosystem (IUCN SSC, 2016).

A proxy species might potentially become invasive, due to genetic factors associated with the proxy species creation process, or factors arising from the rearing environment, or because of ecological and environmental changes since extinction that mean any release might be into an ecosystem where individuals of resident species have never encountered the original form of the proxy. There are also potential adverse disease effects, including the transfer of diseases from captivity (from multiple potential origins) into the wild (always a concern to be addressed in any reintroductions), and impacts on introduced animals of diseases for which they have no prior history of exposure. There is also a small but non-negligible likelihood of endogenous retroviruses being revived along with the proxy species, and thereafter becoming exogenous (IUCN SSC, 2016).

Finally, the status of de-extinct species is complex, and no existing legal framework is entirely suited to such species, and their status under different frameworks is unclear (e.g. Is it still an endangered species? Is it an invasive? Wagner et al., 2017). For this reason, CITES currently has a working group to examine the legal status of "de-extinct" species, and make recommendations for any changes in the Convention or national legislations to ensure that they are both legally protected where appropriate, and also do not undermine legal protections for extant threatened species.

Given the multiple major implications of this concept for biodiversity conservation, IUCN has produced a document of Guiding Principles on the topic (IUCN SSC, 2016). A key, and often overlooked, fact is that species cannot be brought back from extinction; none of the current pathways will result in a faithful replica of any extinct species, due to genetic, epigenetic, behavioural, physiological and other differences. However, synthetic biology, combined with traditional genetic, breeding and husbandry techniques, can theoretically produce proxies of extinct species. The IUCN Guidelines posit that the legitimate objective for the creation of a proxy of an extinct species is the production of a functional equivalent able to restore ecological functions or processes that might have been lost as a result of the extinction of the original species (IUCN SSC, 2016).

5.4 Summary

Certain synthetic biology applications, if appropriately designed and targeted, have potential for enhancing biodiversity conservation. Technological development of such approaches to date has occurred mainly for potential application to the suppression or eradication of invasive alien species. However, no technology developed for conservation purposes is yet ready to be tested in the field, let alone applied for management, with the possible exception of disease resistant American chestnut trees. Application and efficacy of proposed synthetic biology approaches (including gene drive) in the field are likely to encounter multiple hurdles which will require further development to overcome, or may even prove to be intractable barriers to useful application.

Significant concerns exist that genome-editing may cause harm to the individual or population and communities due to uncertainties with altering genome processes and potential subsequent knock-on effects (Lander, 2015). Such concerns arise from the fact that much remains to be learned about how the information that is encoded in the genome is transcribed into function. A further concern is that transgenes or genetic manipulations may horizontally transfer among species, particularly relevant when target species can breed with non-targets. Of greater concern is the potential for synthetic biology approaches intended to be self-disseminating, such as engineered gene drives, to affect non-target populations. Multiple strands of development are thus exploring self-disseminating approaches that are self-limiting or tactically controllable in other ways.

There are also social and regulatory uncertainties surrounding affected communities' interests in having synthetic biology tools deployed. Social science research and stakeholder engagement

will play a critical role in understanding stakeholder values around any potential application of synthetic biology for conservation, and whether the proposed tools are acceptable for use. Should communities and stakeholders support the progression of any synthetic biology approach to field trial stage, it must be borne in mind that consensus on the suitability of field sites for such trials is yet to be achieved. Case-by-case assessments are thus needed of any proposed field sites and trial designs. Such assessments should recognise all potential adverse effects; with the broad uncertainties surrounding impacts to individuals, populations and communities, comprehensive assessments (including ecological) that include contributions from conservation and social scientists and policy experts are critical to any potential deployments of such technology.

Final considerations relate to the 'moral hazard' of resources being removed from more conventional conservation actions and developments to fund investigation into synthetic biology solutions. However, these concerns, and the need to understand the potential adverse effects of the technology, need to be considered alongside the 'moral hazard' of potential species decline and extinction should potential solutions to their loss not be researched and implemented if successfully developed, or their development delayed by an over-emphasis on caution. Thus, such reasons both for and against a role of synthetic biology in biodiversity conservation (including the consideration of the counter factual of what alternative tools and approaches can and could achieve) should always be considered in tandem for robust decision making.

6. Biodiversity conservation implications of synthetic biology applications not directly intended for conservation benefit

Todd Kuiken, Edward Perello, Hilde Eggermont

Image by: Ye Maltsev / Shutterstock.com

6.1 Overview

This chapter examines technology trends and specific examples of synthetic biology tools, applications and products that may indirectly impact conservation goals and threats. While the impacts themselves may be significant, they are indirect because the intended purpose of the technology or application itself is not for conservation. This means that the developers of the product or application have not stated specifically, or it cannot be inferred, that they were developed with the purpose of impacting conservation issues. This assessment does not explicitly address the many concerns and hopes surrounding genetically modified organisms, which has generated a large formal and informal body of literature (NASEM, 2016a).

The chapter begins with a discussion and case studies related to synthetic biology and genome editing techniques for agricultural applications, product replacement, pest management and improving habitat quality. The final section discusses new and emerging issues as well as communities that have access to a suite of technologies related to synthetic biology. These new communities themselves could have an indirect impact on conservation by expanding the community of conservation practitioners, expanding the potential toolbox of options for conservation solutions, and/or expanding the source of adverse effects on conservation.

6.2 Synthetic biology applications for agriculture

Agriculture in the form of croplands and pastures occupy approximately 40 per cent of the Earth's land surface (Foley et al., 2005), making it the planet's single most extensive form of land use (Campbell et al., 2017). Agriculture is also a major factor accounting for biodiversity loss (Chaudhary, Pfister & Hellweg, 2016), ranked second in a global analysis of threats to threatened or near-threatened species (Maxwell et al., 2016). Land devoted to agriculture is also expected to expand in coming decades (FAO, 2016).

Agriculture is one of the major sectors for investment, research and development in synthetic biology. For example, in 2017 Ginkgo Bioworks and Bayer announced an investment of US$ 100 million in a new agricultural biotechnology company (ginkgo-bioworks-bayer-invest-100m-new-agbio-company). Agricultural challenges that are being addressed with synthetic biology and genome editing include climate change (Abberton et al., 2016), soil fertility (Bender, Wagg & van der Heijden, 2016), plant microbiomes (Borel, 2017), photosynthesis (Bourzac, 2017) and crop nutrient content (De Steur et al., 2017). Applications are directed at a wide range of animals and plants (Table 6.1), and are expanding rapidly. To understand the impact of these synthetic biology and/or gene drive approaches a risk assessment would

Table 6.1 Examples of genome editing techniques of relevance to agriculture

Crop/Animal	Type of edit	Results	Reference
Cacao	Gene disruption	Increased resistance to the cacao pathogen *Phytophthora tropicalis*	(Fister et al., 2018)
Cassava	Gene disruption	Increased resistance to cassava brown streak disease	(Gomez et al., 2018)
Cotton	Viral gene disruption	Elevated resistance to cotton leaf curl disease	(Iqbal, Sattar & Shafiq, 2016)
Maize	Promoter disruption	Improved maize grain yield under field drought stress conditions	(Shi et al., 2017)
Rice	Gene mutation	Promoted rice growth and productivity	(Miao et al., 2018)
Rice	Gene disruption	Enhanced resistance to *M. oryzae*	(Wang et al., 2016)

Crop/Animal	Type of edit	Results	Reference
Rice	Promoter disruption	Increased resistance to bacterial blight	(Jiang et al., 2009)
Tomato	Gene disruption	Increased crop yields	(Rodríguez-Leal et al., 2017)
Wheat	Gene disruption	Increased resistance to powdery mildew microcolonies	(Wang et al., 2014)
Japanese black cattle	Gene/cell mutation	Correction of a disease mutation (Isoleucyl-tRNA synthetase syndrome) in Japanese black cattle	(Ikeda et al., 2017)
Pigs	Gene disruption	Increased resistance to Porcine Reproductive and Respiratory Syndrome in pigs	(Burkard et al., 2017)
Poplar trees	Transgenes	Tree sterility	(Klocko et al., 2018)

need to be conducted and compared to alternative approaches (i.e. push-pull agriculture (International Centre of Insect Physiology and Ecology, 2018)).

Intervening in agricultural production systems can have positive or negative consequences for biodiversity (UN CBD, 2015). As with other synthetic biology applications discussed in this assessment, many of these with agricultural relevance are in the early stages of development and clear evidence is not available to properly evaluate their impacts on biodiversity. The potential negative impacts of synthetic biology applied for agricultural purposes on biodiversity and conservation have been discussed by a number of reviews (e.g. Science for Environment Policy, 2016). They include potential impacts such as: transferring genetic material to wild populations through horizontal or vertical gene transfer; having toxic effects on other organisms such as soil microbes, insects, plants and animals; creating new invasive species that may have an adverse effect on native species by destroying habitat or disrupting the food web; facilitating greater application of agrochemicals with biodiversity impacts; reducing soil fertility and structure by allowing for more intensive agriculture; and creating crops that can better utilise marginal land, or even use previously unusable land (Science for Environment Policy, 2016).

Potential benefits to biodiversity include: enhancement of decomposition rates and nutrient fixation (Good, 2018); reduction in the application of fertiliser (Good, 2018); more efficient production of farm animals with concomitant reductions in feed and land use (Van Eenennaam, 2017); forest restoration (Dumroese et al., 2015); and production of livestock feed based on more efficient industrial production of microbial proteins (Pikaar et al., 2018).

6.3 Synthetic biology applications for pest control

Various applications have been proposed using synthetic biology to combat different types of pests, responsible for damage to both agricultural and human health. The case studies below examine two such applications in detail. The first explores the impact of an engineered gene drive approach for malaria vector suppression in Africa, while the second examines a synthetic biology application addressing honeybee colony collapse. While neither of these applications have been released into the environment, other applications, such as a genetically modified diamondback moth, are in the field trial stage (Shelton Lab, 2018). The development of engineered gene drive strategies for malaria vector control and other synthetic biology pest control applications is an emerging field based on modelled population level effects, derived from several different molecular strategies. Many of the plausible pathways to benefit or harm remain hypothetical in the absence of product-specific data to inform case-

by-case risk assessment and manage uncertainty. Those studies largely depend on the final technology developed, its mode of action and unique molecular and phenotypic characteristics. Any application for pest control using synthetic biology or engineered gene drive systems would therefore need to be evaluated against a risk assessment framework (Section 3.4).

Case study 6:

Gene drive approach for malaria suppression in Africa

Delphine Thizy, Luke Alphey

Existing alternatives/baseline situation

Malaria is a leading cause of death in Africa amongst children under five. An estimated 427,000 people died of malaria in 2016 (WHO, 2017). Every year 216 million cases of malaria are reported, 90 per cent of which are in Africa, with an estimated cost of US$ 12 billion for Africa alone (Gallup and Sachs, 2001). Worldwide, human malaria is caused by any of five *Plasmodium* species; in Africa overwhelmingly by one of these, *Plasmodium falciparum* (Snow et al., 2017). The World Health Organisation (WHO) estimates that US$ 9 billion per year would be needed to cover 90 per cent of the population at risk in 2030 with existing malaria prevention and treatment tools; only US$ 2.4 billion is currently available. This investment would significantly reduce but not eradicate malaria.

There are two main types of malaria control interventions in Africa: interventions targeting the disease in humans; and vector control interventions targeting the transmission of the malaria parasite from an infected person to another person through the bite of a female mosquito from the genus *Anopheles*.

Considerable progress has been made towards targeting the disease in humans, however vector-based interventions remain crucial for malaria control. Available vector control tools are diverse, but two have been primarily used in the last decades and have made an important contribution to reducing the number of deaths and infections from malaria: insecticide-treated bed nets and indoor residual spraying (Bhatt et al., 2015). The cumulative impact of those tools on non-target organisms is not well known (Junges et al., 2017). Insecticides may have impacts on non-target species and some formulations, such as those containing DDT, have raised particular concerns in terms of toxicity (Burton, 2009). Despite efforts to end the use of DDT because of its adverse environmental impacts, WHO re-endorsed its use for malaria control in 2006, reversing 30 years of policy (WHO Global Malaria, 2011).

In the past few years, alarming signs of resistance against insecticides have been identified in a number of African countries (http://www.irmapper.com). Research on new molecules is underway to counteract the impact of this growing resistance. Vector control methods need to become more sustainable and more cost-efficient to overcome resistance and advance malaria elimination (WHO, 2015; Killeen et al., 2017).

Current vector control tools also face important challenges in terms of social and cultural acceptance. For instance, there is still a discrepancy between the proportion of bed net ownership and proportion of people

reporting to have slept under a bed net during the previous night (WHO, 2017).

Description of the gene drive approach

Researchers are exploring the possibility of using engineered gene drive mechanisms to suppress the population of *Anopheles gambiae* complex – one of the primary vectors of malaria in Africa – to a point where, in conjunction with other malaria interventions, the cycle of transmission of the parasite could be interrupted and thus contribute to the elimination of malaria. Other engineered gene drive approaches are also being researched to alter the mosquitoes to stop the malaria parasite transmission (Gantz et al., 2015).

The objective is to insert a modification in malaria mosquitoes that would affect the mosquito's ability to reproduce. This could be achieved in two different ways (Burt et al., 2018): by biasing the sex ratio of mosquito populations to have mostly males (only females bite and transmit malaria); or by reducing female fertility (see Appendix 2 (www.iucn.org/synbio) for an in-depth overview of engineered gene drive elements). When introduced in the malaria mosquito, the nucleases work by identifying and cutting essential genes targeted by researchers, such as fertility genes. The interrupted gene will no longer function, and modified mosquitoes will be affected according to the nature and importance of the gene. While there may be some fitness costs in addition to the sterility of female homozygotes, as long as these are not too large the preferential inheritance resulting from the gene drive can ensure the modified gene still increases in frequency over successive generations (Burt, 2003).

The ultimate goal is to produce modified mosquitoes for the malaria vector species that can pass these genes on to a high percentage of their offspring, so the modification is spread throughout the specific target populations relatively quickly and is effectively "self-sustaining" (Burt & Crisanti, 2018). Since malaria is transmitted by several *Anopheles* species, and genetic control tools are highly species specific, any engineered gene drive tool would likely still need to be complemented by other existing approaches (Eckhoff et al., 2017).

As costs and logistical challenges create important limitations on the use of current tools, engineered gene drive approaches, alongside other new tools, could help reach remote areas. They are being thought of in terms of 'first mile' or 'last mile' interventions, where they could help increase the efficiency of existing tools or help achieve elimination in countries that have already significantly progressed but where low-level transmission remains persistent.

Despite progress in the laboratory, any field release for the purpose of evaluating a gene-drive-based construct for vector control is at least a few years away. It is not expected that a fully evaluated vector control technology will be available for another 10 years. This is not only a function of progress in scientific research, which is progressing rapidly, but a function of the large body of knowledge that must be acquired to assess the technique's safety and efficacy. Some remaining areas for technical research are around the emergence of resistance to the editing, which could greatly diminish the efficacy of the tool (Champer, Buchman &Akbari, 2016; Unckless, Clark & Messer, 2016; KaramiNejadRanjbar et al., 2018). This is not unique to gene drive mechanisms but is common for all vector control tools (Kleinschmidt et al., 2018). Researchers working on this technology showed that resistance can arise due to changes in target site caused by the gene drive construct (Hammond et al., 2017). Two ways of retarding resistance have been proposed: (i) targeting multiple sites (Champer et al., 2018), and (ii) targeting conserved sites that cannot tolerate changes while maintaining function. The latter has been demonstrated in small cages (Kyrou et al., 2018). More work is needed to optimise how the two strategies can be best combined to maintain efficacy levels. Additionally, more information

is needed about mosquito populations to feed the models about the dispersion of the genetic modification and to understand if this technology could effectively and safely be deployed for malaria control. For example, there is ongoing research to better characterise the genomic diversity of *Anopheles gambiae s.l.*, which will be instrumental to model the dispersion and understand its potential limitations (Miles et al., 2017).

While there is a societal interest in finding new vector control tools to reduce malaria transmission and to do so in a cost-effective way, as expressed for instance during the Commonwealth Heads of States and Government Meeting in 2018 (CHOGM, 2018), socio-economic and cultural considerations need to be explored for this application of engineered gene drive systems. It is important to ensure that dialogue is taking place with communities, indigenous people and other relevant stakeholders (including research groups) so that their perspectives and values are taken into consideration (Kofler et al., 2018).

Biodiversity and conservation concerns are often raised by stakeholders, as illustrated by the press coverage and as reflected in the call from some organisations for a moratorium on gene drive research. As is the case for all synthetic biology risk assessment (Section 3.4.3), these risks and potential impacts need to be considered relative to the situation without the intervention, i.e. in this case, to the impacts of existing malaria control tools.

Potential opportunities resulting from the approach

The intended direct impact of this approach is clearly the reduction of human malaria (Eckhoff et al., 2017; WHO, 2017; African Union, 2018). However, as engineered gene drive systems would be complementary to other malaria control tools, additional potential conservation benefits could come from the interaction with other malaria reduction tools, for example, the reduction in the use of DDT, which was reintroduced for malaria control in 2006 under certain conditions (WHO Global Malaria, 2011).

An additional indirect impact from the research on engineered gene drive for human malaria control could be to advance scientific knowledge, regulatory frameworks and public engagement for engineered gene drive in mosquitoes. This could in turn benefit other applications of engineered gene drive currently being investigated, notably the use of engineered gene drive for the control of avian malaria (Liao et al., 2017).

Potential adverse effects resulting from the approach

A number of initiatives have started exploring the plausible pathways to harm in order to identify classes of data and information that may be required in order to perform a risk assessment. In 2016, the Foundation for the National Institutes of Health (FNIH) organised a problem formulation workshop to examine hypothetical examples of engineered gene drive applications, including both suppression and alteration strategies, and arrived at consensus points including the following two bullets (Roberts et al., 2017):

- For mosquito biodiversity:

Although this approach aims to target *An. gambiae* in its native range, unlike other case studies where the target is an invasive population, the workshop concluded that *An. gambiae* is not a keystone species and therefore the ecosystem-level consequences of suppression of its populations were unlikely to be severe. Interactions with other species (by feeding on them, being consumed as prey or competing with them) need to be further explored. The toxicity of novel gene products needs to be tested for those interactions as well.

The question of gene flow was considered, and the paper concluded that hybridisation with other *Anopheles* species was likely for some species.

- For animal health:

The potential harm was considered from "pathogen transmission dynamics to livestock." No other relevant pathways were identified, though equivalent impacts on wildlife might be envisioned.

Further to this workshop and publication, the question of gene flow is being investigated by researchers. *Anopheles gambiae s.l.* is a complex of sibling species. Some of these species have on-going gene flow which has been documented (Coluzzi et al., 1979; Fontaine et al., 2015; Neafsey et al., 2015). Therefore, there is a possibility of the gene drive elements spreading by hybridisation and this needs to be considered when assessing risks and benefits. Most of these species from the complex are vectors of human malaria and could be a target for vector control (Bernardini et al., 2017). It is an active field of investigation to see to what extent it is possible to direct gene drive constructs to only one species, despite hybridisation.

Furthermore on the question of the role of *Anopheles gambiae* in the ecosystem, a recent literature review confirmed the statement that it is not a keystone species (Collins *et al.*, 2018). The suppression of *An. gambiae* using engineered gene drive systems may have an indirect effect on conservation through niche replacement – the possibility that another species will fill the now-empty ecological niche previously filled by *An. gambiae* even if those effects could be transient as the aim of those interventions is not extinction but suppression. While this does not seem to have been noted as a problem in control programmes so far, niche replacement may be more feasible in the context of engineered gene drive systems, as their species-specific nature may mean that they do not exclude other species as much as broad-spectrum interventions, such as insecticides, may do. In the specific case of *An. gambiae* it is not obvious that any significant ecological disruption might arise through this mechanism (Collins et al., 2018).

The reduction or elimination of human malaria might lead to demographic and land-use changes, potentially impacting conservation and so should also be considered, although it is of course not specific to engineered gene drive systems and would apply to any successful vectorcontrol intervention.

Case study 7:
Addressing honeybee colony collapse

Daniel Masiga

Issue

Pollinators are essential to food and nutritional security, with about three-quarters of all food crops benefiting from pollination, with the value to global production estimated at approximately US$ 351 billion annually (Lautenbach et al., 2012). Honeybees are critical pollinators of plants, but they are increasingly threatened by pests, pathogens, neonicotinoids and other pesticides (Pisa et al., 2017), and other stressors such as anthropogenic habitat fragmentation. Managed honeybee colonies can be used for honey production, crop pollination, or both. There has been a significant decline in managed honeybee colonies, particularly in Europe and North America (vanEngelsdorp et al., 2009; IPBES, 2016a).

This phenomenon is known as Colony Collapse Disorder, because it is associated with large-scale loss of managed honeybee colonies. Experts believe that multiple factors are responsible for Colony Collapse Disorder, including parasites, like the Varroa destructor mite, bacterial diseases, viral infections and pesticides (van Engelsdorp et al., 2017). Although Colony Collapse Disorder has largely been considered in the context of crop pollination and hive products, significant loss of pollinators can have a large impact on biodiversity and ecosystem services in natural landscapes (IPBES, 2016a).

Existing interventions

In response to the need to preserve honeybee health, the EU recently banned the use of neonicotinoid pesticides (Stokstad, 2018). There are clearly complex interactions between pathogens, pests, pesticides and habitat loss (IPBES, 2016a). Some studies have suggested an association of Colony Collapse Disorder and co-infection with fungi of the genus *Nosema* and invertebrate iridescent virus (Bromenshenk et al., 2010).

Synthetic biology proposed application

Al Dahhan and Westenberg have proposed using a synthetic biology approach, based on the hypothesis that removing either of these pathogens would reduce the occurrence of Colony Collapse Disorder (Foster & Pummill, 2011). The approach proposed is based on the practice of farmers to control one of these pathogens, *Nosema ceranae*, with fumagillin, a compound produced by the fungus *Aspergillus fumigatus*. They propose to engineer a microbe to produce fumagillin, by scanning the fungus genome (Hagiwara et al., 2014) for the pathway responsible forfumagillin synthesis.

Potential adverse effects of the synthetic biology application

These authors concede that differential responses of *Nosema* species (*N. apis* and *N. ceranae*) could render the approach problematic if for example, the use of fumagillin favours the displacement of *N. apis* by *N. ceranae*. It has been demonstrated that *N. ceranae* (and not *N. apis*) weakens the immune system of honeybees, making them more vulnerable to other pathogens (Antúnez *et al.*, 2009). Hence, a significant spread of *N. ceranae* could be damaging to honeybee populations. Such an approach could have negative effects on the natural resilience in honeybee populations. A study carried out in Kenya, where feral honeybee colonies are predominant, has shown that minimally managed honeybee colonies are resilient to stressors associated with colony collapse, such as Varroa mites and a range of pathogens (Muli et al., 2014).

6.4 Synthetic biology applications for product replacement

Synthetic biology has the potential to provide new production methods for new and existing, commercially available products (see: https://www.futurebioengineeredproducts.org/) by changing the production methods and raw material inputs (e.g. petroleum to bio-based). These shifts in inputs could have important positive or negative impacts on conservation. For example, synthetic biology has the potential to replace existing products derived from threatened species (Case study 8) but shifts to a synthetic biology alternative could inadvertently increase the demand for the natural product (Section 5.2.2). Different synthetic biology processes will also be utilised in order to derive these products, which could in turn exacerbate or minimise climate change, land-use change, nutrient cycles and biodiversity loss. Further, global commerce has the potential to translate the production of a synthetic biology application in one part of the world into land conversion in another, such that while beneficial at first glance, an application could generate increased ecological impact if viewed over time and at global scales (Melillo et al., 2009;

Liu et al., 2013, 2015). Analysis of synthetic biology applications may affect the socio-economic impacts on local communities that may be impacted by economic market shifts, which could drive changes in land use and livelihoods, potentially impacting indigenous peoples' cultural heritage as well as conservation. For instance, rising demand for biofuel feedstocks has caused food prices to fluctuate (Westhoff, 2010; Liu et al., 2015).

It is difficult to predict how the dynamics of complex economic systems change when substituting one product for another. The location and choice of organism used as the chassis to produce the new product might also affect the ecosystem dynamics. The following examples were chosen to illustrate how synthetic biology-derived products, at various stages of development, could at least partially replace current products on the market.

Case study 8:

Horseshoe crab replacement for Limulus Amebocyte Lysate test

Tom Maloney, Ryan Phelan

Issue

Three species of Asian horseshoe crab (*Tachypleus tridentatus, Tachypleus gigas* and *Carcinoscorpius rotundicauda*) and the North American species (*Limulus polyphemus*) are all facing global threats. While comprehensive data are difficult to obtain, populations of all four species are currently declining (Vestbo et al., 2018). While the three Asian species of horseshoe crab are considered Data Deficient, in 2016 the American horseshoe crab was re-assessed from Near Threatened to Vulnerable on the IUCN Red List of Threatened Species (Smith et al., 2016). Reduction in horseshoe crab populations have negative impacts on a number of wading bird species that depend on horseshoe crab eggs: six species of shorebirds synchronise their northward migration along the Atlantic flyway to gorge on the eggs of spawning horseshoe crabs in Delaware Bay, a critical food stop on their journey to Arctic nesting grounds (McGowan et al., 2011). The abundance of horseshoe crab eggs is a critical factor to the survival, physical condition and successful breeding of birds, particularly the red knot (*Calidris canutus rufa*), whose 9,500-mile migration from the tip of South America to the Arctic is among the longest of any bird in the world. From 1980 to 2014, red knot populations decreased by as much as 75 per cent in some areas, largely due to the lack of horseshoe crab eggs in Delaware Bay (Mizrahi & Peters, 2009; US FWS, 2014). Fisheries managers now explicitly recognise the interdependence between the horseshoe crab and the migrant shorebirds and have designed a multi-species adaptive management framework to guide management (Atlantic States Marine Fisheries Commission, 2015).

The primary threat to horseshoe crabs stems from their unique role in biomedicine (Krisfalusi-Gannon et al., 2018). Bacterial contamination in the production and delivery of injectable medications and medical devices can cause life-threatening fever or toxic shock if introduced intravenously (Ding & Ho, 2001). Horseshoe crab blood cells known as amebocytes are able to detect minute quantities of endotoxin (molecules present in gram-negative bacteria), and a lysate of horseshoe crab blood, known as the Limulus Amebocyte Lysate (LAL), has become the most

commonly used endotoxin detection method worldwide (Federal Register, 1977). This test, and the necessary collection and bleeding of horseshoe crabs, has been integral to the safe production of vaccines and injectable medications for the past 40 years (Abate et al., 2017), at the cost of severe declines in the species (Smith et al., 2016).

Unlike in Asia, where horseshoe crabs are used for other purposes after being bled (Gauvry, 2015), most crabs in North America are released after bleeding, although some are sold for bait in the whelk and American eel fisheries (Krisfalusi-Gannon et al., 2018). The mortality rate of released horseshoe crabs ranges from 10 to 30 per cent in the US; however, these figures do not account for any further trauma and/or detrimental behavioural changes once the animals are returned to the ocean, nor the derivative population impact from the disruption of horseshoe crab spawning (Krisfalusi-Gannon *et al.*, 2018). The impact of biomedical bleeding on the fishery is compounded by the effects of shoreline development, climate change and rising sea levels, all of which are diminishing the availability of suitable spawning sites (Nelson et al., 2016).

The IUCN Red List of Threatened Species predicts declines of at least 30 per cent in horseshoe crab populations over the next 40 years, while global demand for vaccines, pharmaceuticals and medical devices over approximately the same period will require an increasing supply of LAL. These dynamics create significant uncertainties as to whether current harvesting levels can be sustained, much less meet projected demands (Krisfalusi-Gannon et al., 2018).

Synthetic biology description

The invention of an effective synthetic alternative to the LAL test presents an opportunity for the conservation of horseshoe crabs and the birds that depend on them (Maloney, Phelan & Simmons, 2018). Crab-derived LAL undergoes a series of protein responses in the presence of endotoxin and the first reaction is known as factor C. In the late 1990s, scientists at the National University of Singapore engineered recombinant DNA to replicate the factor C reaction (Ding, Navas & Ho, 1995). This recombinant Factor C (rFC) endotoxin assay was patented and made commercially available in 2003 and eliminates the need to capture and bleed horseshoe crabs (Carmichael et al., 2015). However, whilst rFC has been commercially available for 15 years, a number of perceived factors such as uncertainty over efficacy, regulation, supply chain robustness and industry inertia have limited its adoption.

A recently-published paper summarised the results from 10 peer-reviewed studies that evaluated the efficacy of rFC in the detection of endotoxin in water or therapeutic samples (Maloney, Phelan and Simmons, 2018). Each study demonstrated that commercially available rFC tests detect endotoxins with equivalent or better efficacy when compared to the LAL test. These studies also demonstrate that the commercially available tests meet regulatory requirements (that require the assay to demonstrate as-good or better detection) for replacing LAL for the detection of endotoxins. Notably, pharmaceutical industry experts conservatively estimate that the adoption of rFC only in the testing of water and other commonly used manufacturing materials can result in an estimated 90-per cent reduction in the use of horseshoe crab-derived LAL (Bolden & Mozier, 2018, personal communication, 1 April). This in turn will likely stimulate more widespread adoption of rFC. Patent restrictions have expired, meaning new manufacturers can now begin entering the market – Eli Lilly and Company has already converted to using rFC in three of its major manufacturing facilities – and are increasing the reliability of supply (Bolden, 2018, personal communication, 9 May). It appears that widespread adoption of rFC in the biomedical industry is likely and will remove a significant source of annual mortality to horseshoecrabs worldwide.

6.4.1 Omega-3 oils

Commercial aquaculture has relied on wild-caught fish to provide essential fatty acids to captive stock – in particular docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA). Fishing for these forage fish can impact coastal fish nursery habitats, create local toxicological problems, and put pressure on wild fish stocks and marine food webs (Hites et al., 2004; Domergue, Abbadi & Heinz, 2005). As wild fish stocks decline, commercial aquaculture operators have been challenged by rising costs, responding by reducing their use of fish-sourced oils in feedstock and replacing it with plant oils, leading to a measurable reduction in the nutritional value of farmed fish (Sprague, Betancor & Tocher, 2017; Gasco et al., 2018). Transgenic varieties of DHA and EPA-producing terrestrially-grown crops and microalgae (Abbadi et al., 2004; Woessner, 2004; Adarme-Vega et al., 2012) have been proposed as means of maintaining the quality of fish feed, and reducing pressure on wild fish stocks (Domergue, Abbadi & Heinz, 2005), 70 per cent of which are now at or beyond exploitable limits (Winfield, 2012).

In principal, aquaculture operations could benefit from the cost-effective production of locally-produced synthetic EPA and DHA (Sprague, Betancor and Tocher, 2017) and this could relieve pressure on wild forage fisheries. At the same time the availability of cheaper feedstocks could support an expansion of aquaculture by removing a limiting factor on growth and expanding current impacts on fish nurseries.

Synthetic biology-derived EPA and DHA could potentially impact conservation targets in other areas, depending on the method of production. As EPA and DHA can stimulate growth and reproduction in aquatic invertebrates that may associate with these ponds (Wacker et al., 2002; Arendt et al., 2005; Parrish, 2009), the effects could similarly extend to terrestrial invertebrates, possibly pest species, if the oils are synthesised in transgenic crop fields (Colombo et al., 2018). Insects can retain dietary fatty acids, and an experimentally high DHA and EPA has been shown to have developmental effects on insects (Hixson et al., 2016; Colombo et al., 2018). Even if measures are taken to ensure that the biosynthesis of bioactive compounds occurs only in organs of the plant that are not accessible, for instance the seed, this site-specific production may not prevent a larger animal or indiscriminate feeders from accessing a bioactive compound if it is able to enter the field (Colombo et al., 2018). Growing a synthetic biology product in a field that alters local insects could result in local expansion of the use of insecticides (Colombo et al., 2018). Current knowledge suggests the effects of synthetically-derived compounds will be difficult to predict, be they at the social, economic or environmental level.

Given the challenge of exhaustively testing every possible interaction between a novel genetically engineered plant and a complex ecosystem, industries are considering production systems closed to the environment, potentially enabling less fishing from wild stock, and allowing environmental impact assessments of the transgenic production organism to be more reasonably completed (Sprague, Betancor & Tocher, 2017).

6.4.2 Squalene

Squalane is a cosmetic ingredient that functions as an emollient in lotions and moisturisers and has been used as a softener for more than 25 years, according to the Personal Care Council's Cosmetic Ingredient Review (Personal Care Council, 2003). Squalane is the stable, saturated branched-chain hydrocarbon form of squalene that naturally occurs in large quantities in shark liver oil, other fish oils and in smaller amounts in plants (i.e. olive oil, wheat germ oil, rice bran oil, palm oil). Squalene also exists in humans as a component of sebum, an oily fluid produced by the sebaceous glands (Bergeson et al., 2015).

As shark liver oil contains the greatest yield potential for squalene, the manufacturing process to produce it often involves molecular distillation of shark liver oil and hydrogenation of the distillate, followed by a re-distillation step to produce a purity of about 96 per cent squalene (Bergeson et al., 2015). The use of shark liver oil for cosmetic products is controversial, as a quarter of the world's shark and ray species are listed as threatened (Dulvy, 2014; IUCN, 2014) and shark liver harvest could be

having an impact on their populations. In 2008, the organisation Oceana led a campaign against the use of shark liver oil for the production of squalene, resulting in several cosmetic companies committing to stop or phase out the use of shark squalene in products (Oceana, 2008; McPhee et al., 2014).

Squalene is also found in a variety of plant oils. While the amount of squalene in olive oil is typically less than 0.5 per cent, in 2010 BASF commercialised "plant squalene" derived from olive oil deodoriser distillate, the concentrated waste product from the final step of the olive oil refining process that contains up to 30 per cent squalene (McPhee et al., 2014).

Manufacturing squalene using synthetic biology is an alternative option currently in practice at the biotechnology firm Amyris (Amyris, 2018). Amyris uses synthetic biology to develop synthetic yeast strains that convert sugar to produce β-Farnesene, the natural biosynthetic precursor of squalene (McPhee et al., 2014; Amyris, 2018). The yeast is then removed, followed by a chemical coupling process along with existing hydrogenation and purification technologies that extract and purify the squalane for commercial uses (McPhee et al., 2014).

It is not clear whether sharks are being directly fished for squalene or if they are being harvested for other reasons (Dulvy, 2014). Therefore, the impact of a shift towards a synthetic biology alternative on shark population sizes is not known. Additional data related to market share of companies like Amyris are needed to determine the impacts from the production process (feedstocks), land-use change impacts, and whether there would be any other impacts on shark species by reducing the need for shark liver.

6.4.3 Vanillin

Natural vanilla flavour is obtained from the cured seed pods of the vanilla orchid (ETC Group, 2013). Vanilla is a complex blend of flavour and fragrance, with the most important ingredient being vanillin (Evolva, 2018). An estimated 200,000 people are involved in the production of cured vanilla beans per year, with Madagascar, the Union of the Comoros, and Réunion making up around three-quarters of the worldwide vanilla bean production (ETC Group, 2013). However, because of the cost and supply chain variability of natural vanilla, most products do not use natural vanilla but rather synthetic vanillin made primarily from petrochemicals or chemically derived from lignin (Evolva, 2018). Less than 1 per cent of all vanilla flavour comes from the vanilla bean (Bomgardner, 2016). The biotech company Evolva has developed a genetically engineered strain of yeast that produces vanillin glucoside when fed with glucose (Bomgardner, 2016). While the production of vanillin using synthetic biology techniques appears to be a replacement for the petrochemical production of vanillin, concerns have been raised about the socio-economic impacts to local and indigenous communities who historically have been involved in the production of cured vanilla beans (ETC Group, 2013). Similar concerns have been raised around the production of stevia, in particular issues such as traditional knowledge and access and benefits sharing agreements both recognised under the CBD (Meienberg et al., 2015). These issues are currently being deliberated inside the CBD and will likely impact how/if these synthetic biology applications enter the market.

Other outstanding questions include the potential impact from the feedstocks needed to produce these synthetic biology alternatives, as well as the risks if the containment mechanisms for the production were to fail. Additional market share data on shifts towards vanillin and other flavourings produced using synthetic biology techniques versus petrochemical production will be key to evaluating conservation impacts. Further, the impact from national legislation regarding labelling of products, in particular whether they can be labelled natural or not (Meienberg et al., 2015), and quality of the product, may impact consumer preferences and thus affect the production of vanillin and other flavourings using synthetic biology techniques.

6.4.4 Leather

The raw material used to produce leather is a by-product of the meat industry. Tanners use the hides from slaughterhouses and process them into leather that is used in the manufacture of a wide range of products. The global leather industry uses

approximately 5.5 million tons of raw hides producing approximately 460,000 tons of heavy leather and about 940 million square metres of light leather, including split leather (Joseph & Nithya, 2009). The production process used during the tanning and finishing of leather requires considerable quantities of water and chemicals and can cause significant environmental impacts (Joseph & Nithya, 2009).

Synthetic biology may offer alternative pathways to reducing the biodiversity impacts of leather production. Modern Meadow and Zoa™ is a US-based company that "harnesses the power of design, biology, and engineering to produce the world's first biofabricated leather materials" (Modern Meadow, 2017; ZOA, 2018). Specifically, its technology platform uses DNA editing tools to engineer specialised collagen-producing yeast cells. The cells are optimised to manufacture the type and quantity of collagen required. Once purified, the collagen is formulated and assembled into materials for consumer applications (Modern Meadow, 2017). The yeast fermentation technique requires bio-based feedstocks.

The impact of a synthetic biology alternative to leather, and its impact on conservation, has not been evaluated due to the nascence of the product and a lack of data on market impacts and other factors. A life-cycle assessment of the synthetic biology process will need to be conducted to understand the full impacts on conservation. Key questions include the environmental impacts of the synthetic biology process itself, whether the synthetic biology production method reduces the amount of chemicals currently used in the tanning process of leather goods, the potential impact on the use and disposal of unused animal hides, whether a synthetic alternative to leather will increase the desire and price for natural leather, and the impacts on animal welfare and ranching livelihoods.

6.4.5 Cultured meat

Meat production is a major contributor to global environmental degradation. Currently, livestock raised for meat uses 30 per cent of global ice-free terrestrial land and 8 per cent of global freshwater, while producing 18 per cent of global greenhouse gas emissions (Tuomisto & Joost Teixeira de Mattos, 2011; Alexandratos & Bruinsma, 2012). Livestock production is one of the main drivers of deforestation and degradation of wildlife habitats (Tuomisto and Joost Teixeira de Mattos, 2011) with 34 per cent of the global greenhouse gas emissions related to livestock production caused by deforestation: 25 per cent are methane emissions from enteric fermentation of ruminants, and 31 per cent of the emissions are related to manure management (Tuomisto & Joost Teixeira de Mattos, 2011; Alexandratos & Bruinsma, 2012). Global meat consumption is expected to double by 2050 (Alexandratos & Bruinsma, 2012), potentially doubling the impacts of meat production on the environment (Tuomisto & Joost Teixeira de Mattos, 2011).

Arguments have been made that shifting humans to a plant-based diet could lower the environmental burdens and greenhouse gas emission impacts associated with traditional meat production. One such study found that the substitution of 10 per cent, 25 per cent and 50 per cent of ground beef with plant-based replacements in the US results in substantial reductions in national annual dietary greenhouse gas emissions, water consumption and land occupation (Goldstein et al., 2017). However, increased demand for plant-based proteins also has the potential to increase land-use pressures (Goldstein et al., 2017) and could therefore increase environmental impacts from agriculture practices, depending on where these increased land-use pressures take place.

Alternative sources to conventional and plant-based meat production have been proposed, including using biotechnology and synthetic biology (Servick, 2018). Cultured meat, which is produced by growing animal muscle tissue *in vitro*, might reduce biodiversity conservation impacts relative to conventionally produced meat. Cultured meat can be produced using various genetic tools and techniques, including synthetic biology. Currently, small quantities of cultured meat are produced in laboratories, although large-scale production will require more research (Tuomisto & Joost Teixeira de Mattos, 2011). There are no cultured meat products currently on the market and at least one company is attempting to produce seafood (Carman, 2018; Finless Foods, 2018). Other

meat-alternative products produced, in part, with synthetic biology are available from companies such as Impossible Foods™ (Impossible Foods, 2018).

A study in 2011 found that cultured meat production could potentially emit substantially less greenhouse gas and requires only a fraction of the land and water compared to conventionally produced meat. The study also found that cultured meat could have potential biodiversity conservation benefits by reducing pressure for converting natural habitats to agricultural land. However, the study also suggested that large-scale replacement of conventional meat by cultured meat production may have negative impacts on rural biodiversity and livelihoods due to the reduction in need for – and incentive to maintain – grasslands and pastures (Tuomisto & Joost Teixeira de Mattos, 2011). A separate study which conducted a life cycle assessment for the cultivation of cultured meat found that cultured meat production could require smaller agricultural inputs and land compared to livestock, but those benefits could come at the expense of more intensive energy use (Mattick *et al.*, 2015). The study concludes that "large-scale cultivation of in vitro meat and other bioengineered products could represent a new phase of industrialization with inherently complex and challenging trade-offs" (Mattick et al., 2015). This finding was complemented by Alexander et al. which found that "overall primary energy production was shown to be 46 per cent lower than for beef production, but 38 per cent higher than for poultry meat" (Alexander et al., 2017).

It's not clear if consumers will accept cultured meat as an alternative. Several studies have examined consumer preferences towards cultured meat and found varying responses. A study by Siegrist et al. (2018) found that consumer acceptance could be a major barrier to the introduction of cultured meat because it is perceived as unnatural (Siegrist, Sutterlin & Hartmann, 2018). Indeed, as participants in the study learned more about cultured meat, it increased their acceptance of traditional meat (Siegrist, Sutterlin & Hartmann, 2018). An early study in 2015 found that 9 per cent of participants rejected outright the idea of trying cultured meat, with two-thirds hesitant to try it, and about a quarter willing to try it (Verbeke, Sans & Van Loo, 2015). However, when informed of the potential environmental benefits of cultured meat compared to traditional meat, 43 per cent indicated they were willing to try it and 51 per cent were "maybe" willing to (Verbeke, Sans & Van Loo, 2015).

6.5 Environmental engineering

Loss of habitat is a significant factor in biodiversity loss, affecting a quarter of the Earth's land surface (Pacheco et al., 2018). Restoring ecological values to habitat is of global interest, as exemplified by the Bonn Challenge, with its effort to bring 150 million hectares of deforested and degraded land into restoration by 2020 (http://www.bonnchallenge.org/content/challenge). Restoration efforts have been spotty (Nilsson et al., 2016) and there have been calls for relevant new tools, including those developed by the synthetic biology field (Piaggio et al., 2017). Two areas of environmental engineering that have received some attention are bioremediation and biomining.

6.5.1 Bioremediation

Environmental contamination with inorganic and organic toxicants has increased over the years due to rapid industrialisation, urbanisation and anthropogenic activities. Organic contaminants such as petroleum hydrocarbons, pesticides, agrochemicals, pharmaceutical products and inorganic pollutants such as heavy metals resulting from mining are constantly added to the environment (Wong, 2012). Elimination or mitigation of the toxic effects of chemical waste released to the environment by industrial and urban activities relies largely on the catalytic activities of microorganisms, specifically bacteria (Dvořák et al., 2017). Given their capacity to evolve rapidly, bacteria have the biochemical power to tackle a large number of molecules exposed through human action (e.g. hydrocarbons, heavy metals) or generated through chemical synthesis (e.g. xenobiotic compounds) (Das & Dash, 2017). The development of genetic engineering in the 1980s allowed the possibility of rational design of bacteria to catabolise specific compounds, which could eventually be released into the environment as bioremediation agents (Kellogg, Chatterjee and Chakrabarty, 1981). The complexity of this endeavour and the lack of fundamental knowledge, however, led to the virtual abandonment of such

recombinant DNA-based bioremediation methods only a decade later. Systemic biology, which merges systems biology, metabolic engineering and synthetic biology, now allows the same environmental pollution challenges to be revisited through the use of novel approaches (Dhir, 2017; Dvořák et al., 2017). The focus on contaminated sites and chemicals is now also broadened by the accumulation of plastic waste on a global scale. While plastics such as polyethylene terephthalate (PET) are highly versatile, their resistance to natural degradation presents a serious, growing risk to fauna and flora, particularly in marine environments (Thevenon, Carroll & Sousa, 2014).

The remediation or treatment of contaminants by conventional methods (both physical and chemical) is a costly, time-consuming, invasive approach and causes environmental degradation (US EPA, 1999; Ghana EPA, 2003). For example, to abate acid mine drainage, companies often seal off the contaminated sites or erect barriers to contain the acidic fluids (Klein *et al.*, 2013). In order to remediate acidic effluents in the polluted area, chemical treatments, such as the use of calcium oxide that neutralises the acid, are typically applied. To inhibit the acidophilic microorganisms responsible for the acid generation, certain organic acids – sodium benzoate, sodium lauryl sulfate or quaternary ammonium compounds – are used. Many of these treatments are complicated and expensive to apply (Jerez, 2017).

There is now a portfolio of systems-level metabolic engineering tools applicable for biodegradation purposes (Dvořák et al., 2017), providing an alternative to more conventional techniques. These tools are used to gain deeper insight into the genetic and physiological background of the target organisms, to model enzymatic reactions and to determine the constraints for efficient biocatalysis. For example, Austin et al. (Austin et al., 2018) have characterised the 3D structure of a newly discovered enzyme that can digest highly crystalline PET, the primary material used in the manufacture of single-use plastic beverage bottles, some clothing and carpets. They engineered this enzyme for improved PET degradation capacity and further demonstrated that it can also degrade an important PET replacement, polyethylene-2,5-furandicarboxylate, providing new opportunities for biobased plastics recycling.

Despite the clear progress of biochemical and biological engineering in the last decade, the vast complexity of the living cell remains the major hurdle for developing synthetic biology approaches (Dvořák et al., 2017). In the case of biosensing, biodegradation pathway design and the prospective applications of the genetically-modified microbes, the complexity of intercellular and interspecific interactions, and the interplay between the biotic and abiotic factors that govern contaminant biodegradation in polluted ecosystems, are still poorly understood (de Lorenzo, 2008; Meckenstock et al., 2015). Basic events need to be understood just as much as the adverse effects, which also must be identified and assessed as the technology advances.

6.5.2 Biomining

Mining activities have been carried out for thousands of years and currently supply important industrial metals, including copper, iron and gold. Although modern mining companies have sustainability programmes that include tailings management and external verifications, it is recognised that these industrial activities are responsible for significant damage to the environment (Jerez, 2017). In particular, technologies such as smelting and roasting generate toxic emissions, including the release of solid particles into the air (Jerez, 2017). Mining operations can produce large tailings which can generate acid mine drainage (AMD) that affects both environmental and human health (Jerez, 2017). Consequently, and due in part to environmental laws and regulations, these methods are being replaced, in countries such as Chile, Brazil, South Africa and Australia, by less contaminating processes, such as biomining (Harrison, 2016).

Biomining is a generic term used to describe the utilisation of microorganisms to process metal-containing ores and concentrates by bioleaching and biooxidation (Brune & Bayer, 2012). Bioleaching is typically used in the extraction of base metals, where the metals of interest are solubilised through microbial action and are recovered from solution. Biooxidation is generally used for the pre-treatment

of recalcitrant gold and silver bearing minerals, where the microorganisms are used to oxidise the mineral sulfide matrix in which the metal of interest is located. After the undesirable sulfides are dissolved from the minerals, the gold or silver is typically leached with chemical lixiviants, such as cyanide. Both bioleaching and biooxidation utilise similar acidophilic iron and/or sulfur-oxidising microorganisms to solubilise metal containing sulfides. Although biomining offers an economically viable and cleaner option, the acidophilic microorganisms mobilise metals and also generate AMD, potentially causing environmental harm. The same microbes and groups of microbes called consortia that are used in biomining operations are thus the major contributors to AMD generation (Brune & Bayer, 2012).

There is also an increasing interest in applying biomining technology for leaching metals from low grade minerals and wastes. In such cases, however, bioprocessing is often hampered by the presence of inhibitory compounds that originate from complex ores (Gumulya et al., 2018).

One company plans to use synthetic biology to develop microbes to extract copper more efficiently from the ore (Bergeson et al., 2015; Universal Bio Mining, 2018). These novel microorganisms will be designed to increase the solubility and extraction of copper from ore that, using current technology, either could not be extracted or could not be extracted in a cost-effective manner. The company plans to change the microbes by modifying the genetic material to increase the microbes' efficiency in leaching specific types of low-grade ore and may seek to use the modified bacteria to recover additional copper from tailings. The leaching system occurs in a loop. Once the primary copper extraction is complete the remaining leachate is reinoculated with microbes and reintroduced at the top of an ore heap rather than being disposed and potentially contributing to environmental contamination. Because of the routine addition of new inoculant, the microbes are not engineered for maximum stability and fitness and indeed cannot survive at more neutral pH (>3) (Bergeson et al., 2015).

Gumulya et al. (2018) have reviewed the state-of-the-art tools to genetically modify acidophilic biomining microorganisms. They also reviewed the limitations of these tools – both with regard to resilience pathways that can be engineered in acidophiles to enhance their robustness and tolerance in harsh environments that prevail in bioleaching, as well as with regard to the efforts that have been carried out towards engineering robust microorganisms and developing metabolic modelling tools. They explain that – despite a number of complete genome sequences being available for biomining species – only a handful of genetic modifications have been reported. They also show that at present, no genetically modified organisms are being used in commercial scale biomining, and that some heterologous expression vectors and markerless gene replacements have been developed for biomining organisms, albeit with limited efficiency.

6.6 Changing innovation frontiers in synthetic biology

Many new tools and processes from synthetic biology are under development or are on the horizon. Some could have clear relevance to conservation, even if they are still in the conceptual stage, while others will have less obvious consequences for conservation. In both cases, for the purposes of this assessment it is important to have as full an understanding as possible of the trajectory of research and innovation in synthetic biology that might impact conservation. This section provides a broad look at five developments in the field: digital sequence information; reverse engineering genomes for discovery; the International Genetically Engineered Machine Competition (iGEM); the Biodesign Challenge; and DIYBio. The first two showcase changing tools which may enable the collection, storage and sharing of data from the environment, and potentially enable more complex experiments in the laboratory. The last three examples describe a potential expansion of access and interest, especially among young people, in synthetic biology which could impact future innovations, or consequences, for conservation. While it's unclear what those impacts may be, new collaborations from a diverse set of players in environments that nurture imagination have a potential to impact conservation. Whilst conservation has been a motivator for some of these new collaborations or actors it is difficult to assess the extent to which these collaborations

have fulfilled that potential and whether they will make a measurable impact on conservation goals.

6.6.1 Digital sequence information

Digital sequence information (DSI) is the product of DNA, RNA and protein sequencing technologies. Generally these have become faster, cheaper and more accurate in recent years allowing for computational analyses and simulations (Wynberg & Laird, 2018) that previously were unavailable or required cumbersome laboratory experiments. The use of these technologies poses a governance challenge (Section 2.3.2).

Broadly, sequencing technologies used to produce DSI are designed to determine the order in which each of the four nucleotides in a DNA molecule are arranged (Wynberg & Laird, 2018). Sequencing technologies have evolved rapidly, giving rise to next generation sequencing, deep sequencing or high throughput sequencing, making it possible to sequence entire genomes or sample entire transcriptomes more efficiently and in greater depth (Wynberg & Laird, 2018). Sequencing technologies have led to vast amounts of data being produced giving rise to the need for bioinformatics – computational tools and software that enable the storage, analysis and manipulation of large biological datasets, leading to capabilities like metagenomics (Wynberg & Laird, 2018).

Metagenomics enables researchers to sequence and analyse gene sequences from environmental samples; for example, microorganisms and/or invertebrates present in a sample of soil or water. While whole genome sequencing describes the genome of one specific organism, metagenomic analysis produces data from millions of small fragments of the genome of each organism in the sample (Laird & Wynberg, 2018). DNA barcoding can act as a genetic fingerprint by focusing on genes that are present in most organisms, but are also unique to each species. While not applicable to all species, this technique can allow for rapid species identification if databases of sequences are available for comparison (Hebert, Cywinska & Ball, 2003; Conservation X Labs, 2017; Wynberg & Laird, 2018).

Continuous innovation in DNA sequencing approaches has allowed a major increase in the scale, and decrease in cost of applying classical genetics to conservation, to fill gaps in biodiversity knowledge (DeSalle & Amato, 2004, 2017). The practice of working with whole genome datasets is likely to become routine in coming years (Fuentes-Pardo & Ruzzante, 2017). As costs continue to drop and new affordable and accessible tools become available (Conservation X Labs, 2017), DSI and dematerialisation could greatly influence conservation practices and programs (Wynberg & Laird, 2018).

DSI presents conservationists with new capabilities for measuring and acting to minimise loss of genetic diversity (Ba et al., 2017; Hou et al., 2018), understanding population structures of endangered species (Miller et al., 2011; Niissalo et al., 2018), defining pedigrees and cryptic species as management units (Niissalo et al., 2018), and monitoring impacts from human development (McCartney-Melstad, Vu & Shaffer, 2018) to name a few.

Overall the hardware, software and wetware of modern DSI approaches is supporting acquisition of overwhelming volumes of data that can be used for conservation practice. Exemplifying this is an ambitious effort to sequence Earth's Whole Genome (Lewin et al., 2018), which could provide conservationists with digital reference material to potentially make high resolution assessments of biodiversity. DSI's expanded use could support direct measurement of the impact of policies and actions by governments, companies and organisations on biodiversity goals. Nonetheless, like other genomics technologies with the potential to make an impact on the field, conservationists must address many of the gaps in infrastructure, skills and funding to support routine use (Shafer et al., 2015), as well as the socio-economic, cultural and access and benefits sharing impacts of increased access/use of digital sequence information.

Box 6.1
Earth Biogenome Project

In November 2015, a group of biologists proposed a plan to sequence all eukaryotic organisms (animals, plants, algae and fungi are all eukaryotes) on the planet (*The Economist*, 2018). This plan has since developed into the Earth Biogenome Project which was officially announced at the 2018 World Economic Forum in Davos. The goal of the project, estimated to cost US$ 4 billion, is to sequence within 10 years the genomes of all known species of eukaryotes (*The Economist*, 2018; World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018). The scale and complexity of the project is not lost on its developers, and they state they will "rely on convening multi stakeholder collaborations that draw in science, research, technology and ethics communities, along with governments and the private sector" (World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018).

The question this project is asking is: "Could genome sequencing be harnessed to unlock nature's biological inheritance, honed by evolution over millennia?" (World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018). This is hugely ambitious, given that only 14 per cent of plant and animal species on land have been described to science (Mora et al., 2011), and less than 0.1 per cent of those have been sequenced (World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018), and access and benefit-sharing agreements codified in treaties like the Nagoya Protocol and ITPGRFA will need to be addressed (Section 2.2.4).

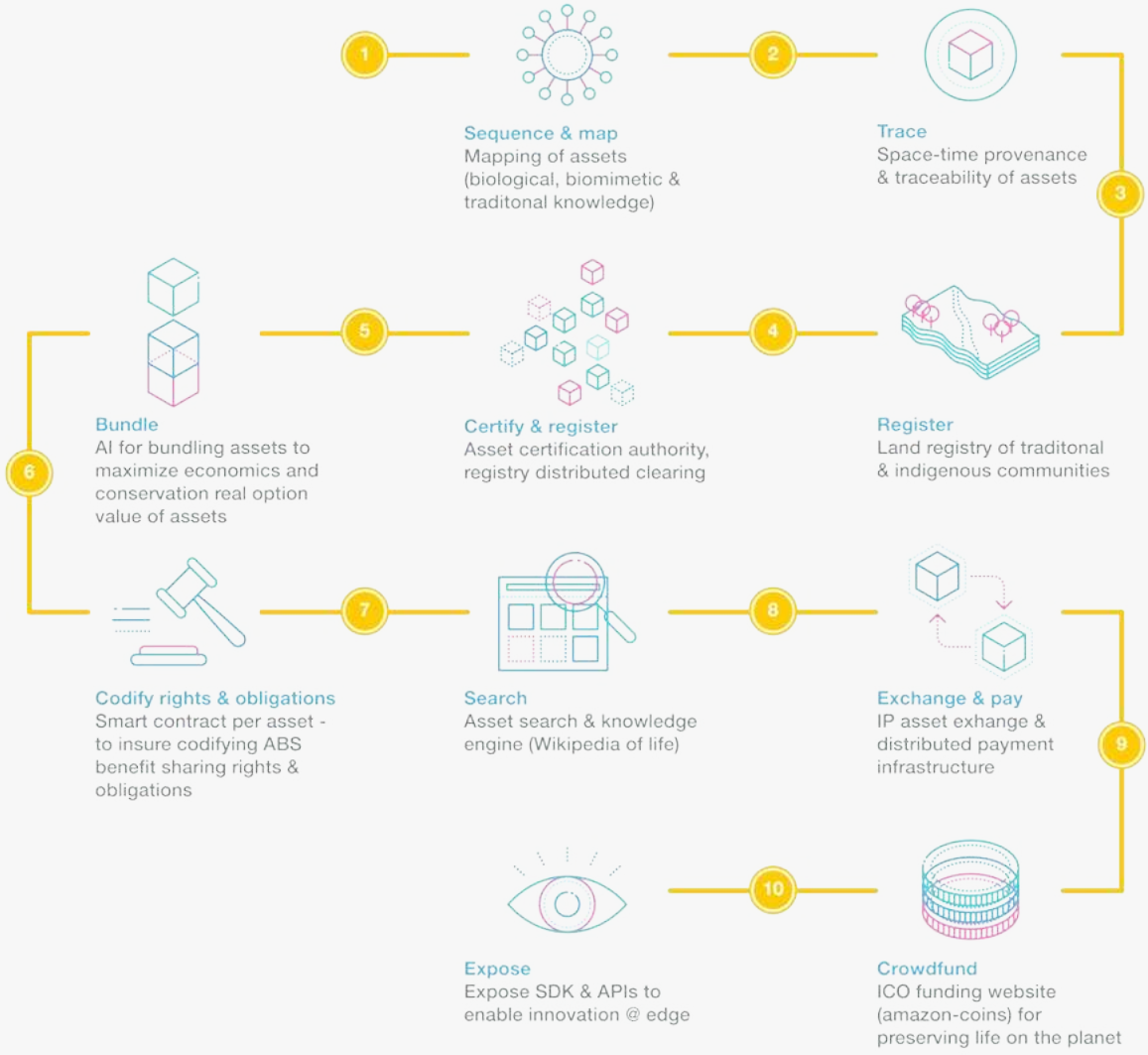


Figure 6.1 The Earth Bank of Codes Platform Structure. Adapted from World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018.

The Earth Biogenome Project plans to utilise blockchain technologies in order to store and track the access to and subsequent usage of the digital information that will be generated in what they are calling the Earth Bank of Codes (Figure 6.1) (World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security, 2018). A blockchain is a digitised, decentralised, public ledger typically managed by a peer-to-peer network that follows an agreed upon protocol. Both Nagoya and ITPGRFA are currently deliberating over whether digital sequence information meets the definition of genetic resources and this will need to be settled before blockchain could be implemented. Whether or not they will experiment with the use of blockchain technology to manage issues of access and benefits sharing is an open question.

6.6.2 Reverse-engineering and understanding genomes

Often the potential for forward engineering and "rationally-designed organisms" overshadows technical conversations about synthetic biology. But for such projects to exist, scientists must know exactly what genes to engineer, and how. This can be determined by systematically modifying an organism to build substantive knowledge of *normal* function, in a kind of reverse engineering, or functional genomics. Much of the biotechnology field has deployed tools and processes associated with synthetic biology to answer the question "how does an organism work," often with humans and their mouse models in mind. However, the technology and the knowledge derived from its use could also answer important questions facing conservation.

Genome perturbation is one such process that embodies a reverse engineering approach. By leveraging programmable nucleases, like CRISPR-Cas9, it is possible to modify genes methodically in order to discover their function. Edited experimental cells and organisms, contained to the laboratory, can be exposed to any number of chemicals or environmental stresses to understand how certain gene variants are relevant to a particular trait. Follow-up studies can then confirm the gene-trait relationship using a more deliberate forward engineering approach and experimental assay. A computational and automated approach to this process lends itself to screening many millions of variants simultaneously, supporting the rapid identification of potential interventions for conservation approaches (assuming the cell biology capabilities are also available for the organism in question). The true power of these genetic manipulation techniques, whether applied in small or large scales, is their potential to directly confirm causal relationships rather than using more limited computational methods to infer causation (Meinshausen et al., 2016). This is useful not only for identifying cause-effect relationships of a conservation-relevant problem, but also solutions. For instance: identifying potential treatments for diseases with no known cure like white-nose syndrome in bats (Cheng et al., 2017), identifying susceptibility and resistance traits to blights and their plant hosts (Lan et al., 2008), or directly confirming how disruptions in the microbiome affect amphibians (Bates et al., 2018).

Reverse-engineering activities for discovering gene and genome function can produce impressive knowledge about biological systems, which could in turn inform conservation science and action. The technical capacity exists, as proven by the application of biotechnology to medicine, but for conservation applications, for *discovery-for-conservation* programmes to lift off, there will be a need for funding, personnel and technical infrastructure (Shafer et al., 2015).

6.6.3 iGEM

Synthetic biology is multidisciplinary, with the most represented disciplines including biochemistry, cell biology, genetics, computer science, engineering and computational biology (Shapira, Kwon & Youtie, 2017). Many of these disciplines are themselves associated with open collaborative movements, which synthetic biology has been inspired by, and draws upon. Today, an active system of public laboratories, community projects, citizen science enterprises and public competitions use synthetic biology approaches. The capabilities of these groups are diverse, and the nature of their work lies along a spectrum of tinkering to engineering, depending on the degree to which a project has been planned (Keulartz &

van den Belt, 2016) and the nature of the tools and experience available to each individual or group. Generally speaking these groups have benefited from a combination of low-cost enabling technologies, the commoditisation of key reagents like synthesised DNA, and the culture of synthetic biology that is open to multidisciplinary projects (Redford et al., 2014).

The International Genetically Engineered Machines competition (iGEM) is an annual synthetic biology event where undergraduates, graduates, high school students and community biotech labs (DIYbio) compete to build genetically engineered systems using standard biological parts called BioBricks. According to the Registry of Standard Biological Parts, which is maintained by the iGEM Foundation, a BioBrick or a biological part "is a sequence of DNA that encodes for a biological function, for example a promoters or protein coding sequences. At its simplest, a basic part is a single functional unit that cannot be divided further into smaller functional units. Basic parts can be assembled together to make longer, more complex composite parts, which in turn can be assembled together to make devices that will operate in living cells" (IGEM, 2017).

Teams are provided with an initial kit that contains about 1,700 parts, and throughout the competition, they create new parts and improve other parts contained in the registry. All these parts are available for anyone to access, use and share. There are over 20,000 documented genetic parts in the Registry and "teams and other researchers are encouraged to submit their own biological parts to the Registry to help this resource stay current and grow year to year" (IGEM, 2017).

iGEM began in January 2003 as an independent study course at the Massachusetts Institute of Technology (MIT) where students developed biological devices to make cells wink on and off. This course became a summer competition with five teams in 2004 and continued to grow to 13 teams in 2005; it expanded to 340 teams in 2018, reaching 42 countries and over 5,000 participants. Since 2004, over 40,000 students have participated in iGEM from across the globe (Figure 1.6 and Figure 6.2). Team projects have ranged from simple biological circuits to developing solutions to local and global environmental conservation issues.

In 2016 the team from the University of Wageningen in the Netherlands designed a synthetic biology system

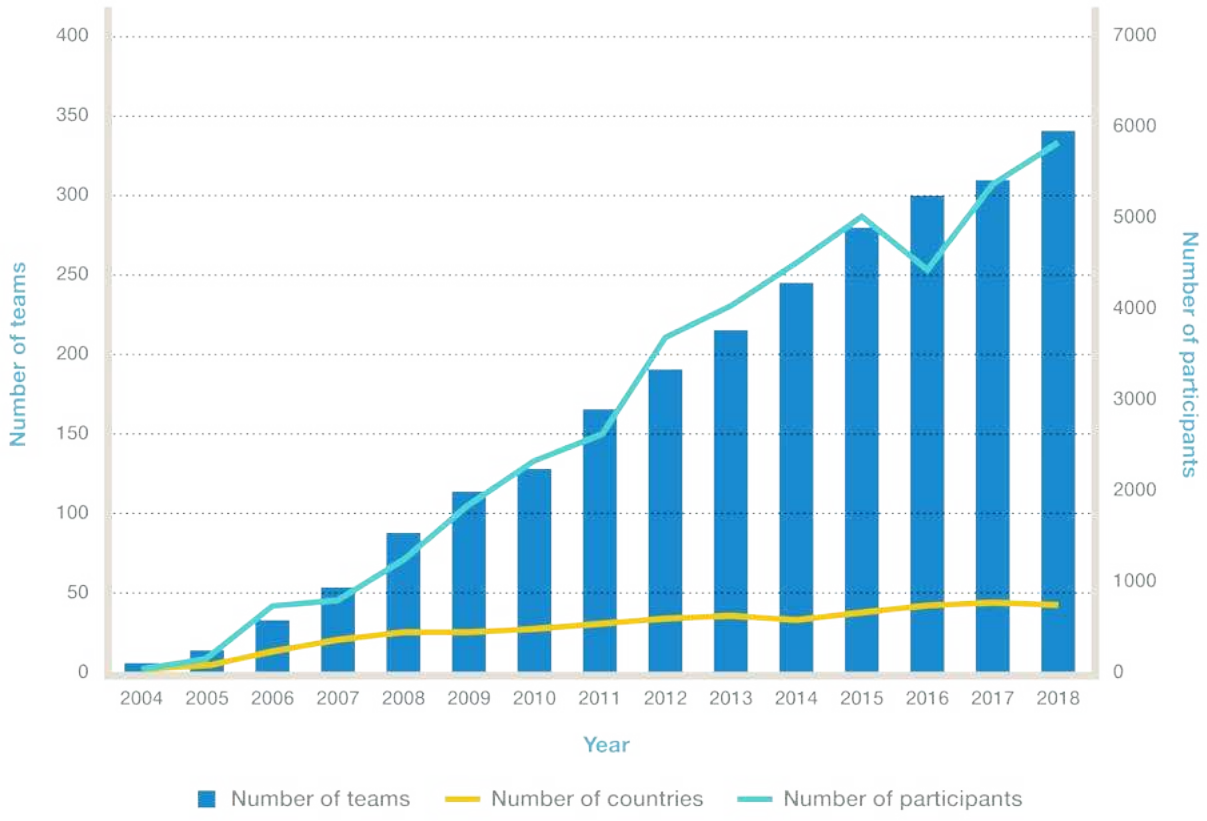


Figure 6.2 Global participation in iGEM from 2004–2018. Adapted from iGEM, 2018.

to address bee colony collapse (Team Wageningen, 2016). Also in 2016, the team from the Federal University of Amazonas and the Amazonas State University developed a project to address mercury contamination in the Amazon basin (UFAM-UEA_Brazil, 2016). iGEM places as high a priority on students learning the technical skills of synthetic biology as it does on them understanding and contextualising how 'human practices' (IGEM, 2018) will influence the impacts of their technology, and how to best plan for potential consequences. Through the human practices component of iGEM, teams are required to study "how your work affects the world, and how the world affects your work" by imagining their projects in a social/environmental context and engaging with communities outside their lab to better understand issues that might influence the design and use of their technologies. To address safety and security issues associated with projects, iGEM has established a safety and security committee which evaluates every team's project at various stages of development. Teams are required to submit check-in forms and subsequent approvals are needed depending on the type of project being proposed (iGEM, 2017). With tens of thousands of graduates from throughout the world iGEM could provide a ready-made pool of people with skills to help conservation if it decides it wants to develop new synthetic biology approaches.

6.6.4 The Biodesign Challenge

The Biodesign Challenge (Biodesign Challenge, 2018) is an annual art and design competition that offers opportunities to university art and design students to develop projects around potential biotechnology applications, some of which directly or indirectly relate to conservation. Students are connected with a team of biologists and experts to guide them as they develop their ideas. At the end of the semester teams showcase their designs in front of members of the academic, industrial and design communities. The competition is based upon a theory that design plays an integral role in the development of any technology and that a designer's vision can both anticipate and inspire new applications which in turn can drive the scientific community and influence society's preferences around technologies (Biodesign Challenge, 2018). These principles have expressed themselves in a number of Biodesign Challenge projects with implications for conservation. In 2017, the New York University team connected beekeeping, synthetic biology and conservation by using modified baker's yeast to produce beta acids that target the parasitic bee mites that contribute to bee colony collapse (NYU Biodesign Challenge, 2017). Other teams have focused on developing biodegradable materials to replace non-biodegradable counterparts, or have worked on biosynthetic alternatives to animal-sourced textiles (Sullivan, 2018). The Biodesign challenge has notably received sponsorship from both People for the Ethical Treatment of Animals (PETA) and the Stella McCartney Foundation, which cite their desire to see a biofabricated wool as motivating their support for the competition (Sullivan, 2018). The involvement of fashion colleges in the Biodesign Challenge is noteworthy, and can be seen as a response to the fashion industry's desire to source sustainable textiles and materials (Kerr & Landry, 2017), especially those that could replace wool, leather and silk, which have major environmental impacts on water scarcity, resource depletion and eutrophication (Higg Materials Sustainability Index, 2018), not to mention land use.

6.6.5 DIYbio

Do-it-yourself biology, or DIYbio, is a global movement spreading the use of biotechnology and synthetic biology tools beyond traditional academic and industrial institutions to other publics (Grushkin et al., 2013). Practitioners include a broad mix of citizen scientists, amateurs, enthusiasts, students and trained scientists, some of whom focus their efforts on using the technology and knowledge to create art, explore biology, create new companies or simply to tinker. Others believe DIYbio can inspire a generation of bioengineers to discover new medicines, customise crops to feed the world's exploding population, harness microbes to sequester carbon, solve the energy crisis, or even grow our next building materials. Whether or how this growing community of biologists, and the expanding access to tools related to synthetic biology, will impact conservation is an open question.

The concept of amateur biotechnologists – what eventually became DIYbio – began to take shape around 2000, after a working draft of the human

genome was completed by the Human Genome Project (Grushkin, Kuiken & Millet, 2013). People began setting up home labs (Carlson, 2005), which evolved into dedicated labs in commercial spaces. The organisers pooled resources to buy, or take donations of, equipment, and began what have become known as "community labs." The first opened in the US in 2010. These labs sustain themselves on volunteers, membership donations and paid classes. DIYbio continues to grow rapidly. There are now community laboratories and other types of community biotech incubator spaces spread across six continents (Figure 6.3). They participate in iGEM, provide educational opportunities and are actively being sought after for innovation opportunities in the conservation arena (Conservation X Labs, 2018).

The Citizen Salmon project (SoundBioLab, 2018), for example, based at the Seattle community laboratory SoundBio, was set up to apply synthetic biology techniques to develop a database of salmon genotypes, and create a DIY genotyping kit for citizen scientists to determine the origin of their store-bought fish (Martin, 2017). The project was notable as a DIYbio initiative that had the potential to advance conservation of king salmon without having conservation as an explicit goal (Martin, 2017). The project highlights the possibility of interactions between citizen scientists, the synthetic biology field, and the development of conservation tools; as costs in enabling technologies decline there is the potential for additional projects of this nature to emerge.

The DIYbio community believes that wider access to the tools of biotechnology, particularly those related to the reading and writing of DNA, has the potential to spur global innovation and promote biology education and literacy that could have far-reaching impacts – and it raises valid questions about risk, ethics and environmental release for all scientists, policymakers and the public (Kuiken, 2016). For instance, Odin, a company that believes "the future is going to be dominated by genetic engineering and consumer genetic design" creates "kits and tools that allow anyone to make unique and usable organisms at home or in a lab or anywhere" (Odin, 2018). Some of these kits raise serious environmental and ethical issues regarding animal welfare (Bloomberg, 2018), along with societal questions about who should be able to access these technologies.

The distributive and democratised nature of synthetic biology techniques presents both opportunities and challenges for the conservation community.

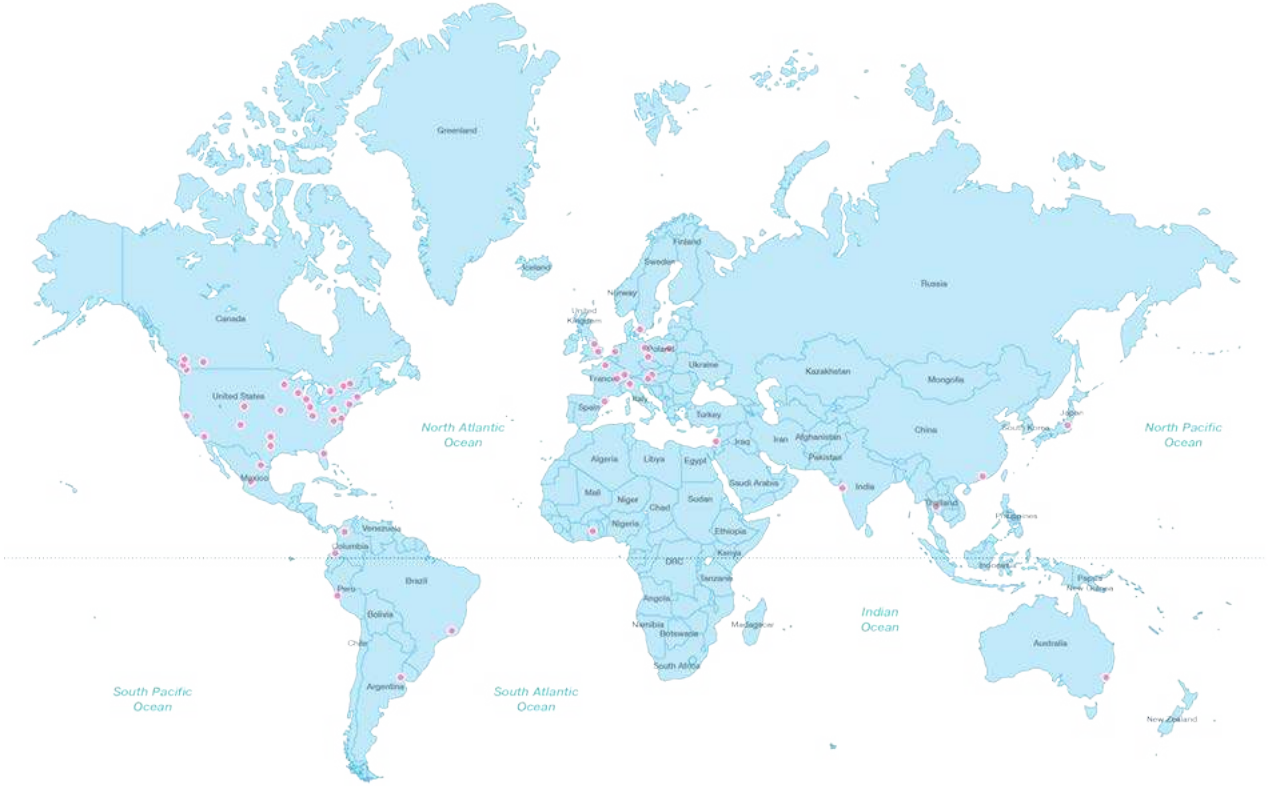


Figure 6.3 Map of community biotech labs and community incubator spaces as of 2018. Adapted from http://sphere.diybio.org/ and personal communications.

7.
Summing up and looking forward

Kent H. Redford, Thomas M. Brooks, Nicholas B.W. Macfarlane, Jason Delborne, Jonathan S. Adams

Image by: Independent Birds / Shutterstock.com

7.1 Synthesis

This assessment provides an opportunity for IUCN Members to consider the evidence regarding the potential positive or adverse impacts of synthetic biology on the conservation and sustainable use of biological diversity. The assessment is rooted in the fundamental idea that decisions on the use of technology need to be informed by empirical studies examining their efficacy, potential benefits and risks. At the same time, the members of the Technical Subgroup responsible for the assessment were keenly aware that, given the nascent stages of most synthetic biology application for conservation, questions of how to address the issue of uncertainty are critically important (Figure 3.1). The assessment's review of the tools of synthetic biology – which, as introduced in Chapter 1, include the concept of engineered gene drive systems – and their potential applications to conservation, and the accompanying case studies (Chapter 4–6) illustrate the broad range of scales at which synthetic biology and conservation may intersect, from small islands to all of sub-Saharan Africa. Such diverse potential both reinforces the need to ground any decisions about the future of synthetic biology and conservation on scientific evidence and offers important context for the debate. The assessment was done with a focus on scientific evidence and hence dictated the expertise of the Technical Subgroup, but this should not be taken as dismissing the role of traditional knowledge, religion and ethical values in decision making. These other types of evidence and other ways to examine risk and opportunity must be considered, and some communities, such as the Māori of New Zealand, are already proceeding with their own analysis (Mead, Hudson, & Chagne, 2017).

Conservation organisations and conservation scientists have long understood that the most significant threats to biological diversity relate to changes in the way humans use land, water and oceans, and also the species they contain. An analysis of the IUCN Red List of Threatened Species (Maxwell et al., 2016; IUCN, 2018) confirms that overexploitation of species and the expansion and intensification of agriculture inflict by far the most significant pressure on threatened or near-threatened species worldwide. Loss of intact ecosystems through destruction and degradation are also major threats and climate change exacerbates all these threats. Such loss also negatively affects the sustainable use of biological resources. The landscape of conservation threats is clear.

For many in conservation the tools to address those threats are clear as well. Decades worth of conservation work has produced some major successes – such as the recovery of whales and the conservation of species through well-designed and funded protected areas – yet there is a clear sense that the threats are getting worse and that current tools may not be able to address emerging threats. Therefore, some conservation scientists have a strong desire to explore the exploding field of synthetic biology, looking for ways that it might be able to help conservation address these intractable problems. There is also an incipient interest in engaging with the synthetic biology field to think of ways that new technologies might produce environmental benefits. At the same time there is deep concern in some parts of some societies that applying synthetic biology tools to environmental questions is an undertaking fraught with uncertainty and potential threat.

This assessment emerged, in part, from concern and from hope among broad segments of society and from a broad discussion that is taking place throughout the world on the proper place of synthetic biology in societies and in nature. It is based on the fact that the communities of conservation scientists and synthetic biologists have operated largely in isolation from one another but that that isolation will not remain. While investment in synthetic biology is expanding rapidly (Figure 1.7), little of that investment is directed at applications intended for specific conservation benefits. The bulk of effort remains on products and processes that may improve agriculture (i.e. more disease-resistant or productive crops or livestock) or human health (i.e. new medicines or approaches to diagnosing or treating diseases and preventing their transmission). So, a key question that emerges is where these areas of effort overlap with conservation and sustainable use, and what the intended and unintended impacts will be on biodiversity. As seen in Chapter 5, the tools and techniques of synthetic biology may be useful in addressing conservation challenges such as invasive alien species, wildlife trade and disease, although with

the potential for adverse effects as well. At the same time, as seen in Chapter 6, efforts are underway to change the production methods and raw materials needed for products like Omega-3 oils, vanillin and others. There is the potential for synthetic biology to develop new techniques to solve such problems as invasive species on islands or chytrid fungus, but at the same time to develop mechanisms that may change land-use patterns in ways that may be harmful or beneficial to biodiversity – or both. Each of these cases will need to be assessed on its own merits, as no technology can be applied universally.

The evidence necessary to provide unequivocal answers to questions about the relationship between synthetic biology and conservation does not yet exist. Deeper collaboration between conservation scientists and synthetic biologists will be necessary to both develop evidence and to create the frameworks for understanding and using that evidence. Scientists are also not the only voices; society needs to be involved and may decide that some research should not proceed, in which case there will be no new evidence. It is already clear, however, that the opportunity to shape how these fields interact and to set the research agenda is here now and will require the engagement not just of scientists but also government at all levels, civil society and indigenous peoples' organisations worldwide.

Key Messages

1. **Synthetic biology and engineered gene drive have important implications for the conservation and sustainable use of biological diversity {1.1, 4.3} that are both direct {5} and indirect {6} (*well established*)**. While most synthetic biology and gene drive products are not designed as conservation applications {1.6} (*well established*), some of these will nonetheless have substantial impacts on conservation practices and outcomes {6.1} (*established but incomplete*).

2. **New tools are needed for effective conservation and sustainable use of biological diversity {1.1} (*well established*).** In recent years, global, regional and national measures promoting biodiversity conservation have resulted in some successes, but biodiversity continues to decline globally {4.3} (*well established*). Biodiversity conservation requires the continued application of proven approaches, but scaling these efforts up to the level necessary to reverse the declines will continue to be a major challenge, given the seemingly intractable nature of some of the threats {5.1} (*well established*). Some synthetic biology and engineered gene drive applications, if appropriately designed and targeted, could enhance biodiversity conservation, for example, by mitigating threats {5.2} and increasing species' resilience to them {5.3} (*speculative*).

3. **The practice of synthetic biology is increasing rapidly, with major developments being promised and some delivered across multiple sectors {1.6} (*well established*).** Over the last 15 years there has been a five-fold growth in companies with public and private investment approaching US$ 10 billion over this period {1.6} (*established but incomplete*). Synthetic biology labs are found throughout the world in academic, corporate and non-traditional spaces like community biotech labs; increasingly young people are being taught to use these technologies {6.6} (*well established*). The distributed nature of access to synthetic biology techniques (*well established*) presents both opportunities and challenges for the conservation community {1.6, 2.3, 6.6} (*speculative*).

4. **Engineered gene drive systems can be a transformative tool for direct conservation applications {5.2.1, 5.3.1} (*speculative*) as well as in other sectors like public health {6.3} (*speculative*), where they could have an indirect impact on conservation {5.2.1, 5.3.1, 6.3}.** Engineered gene drive systems are still years away from any deployment {5.2.1, 5.3.1, 6.3} (*established but incomplete*) despite the fast pace at which this technology is being developed {1.4} (*competing explanations*). The expertise of the conservation community is vital to the responsible development and deployment of engineered gene drive systems {5.2.1, 5.3.1, 6.3} (*well established*).

5. **Synthetic biology and engineered gene drive may be beneficial to conservation and sustainable use of biodiversity {4–6} (*speculative*).** For example, by protecting threatened species against disease or climate threats {5.3.1} (*speculative*), eradicating invasive species {5.2.1} (*speculative*), increasing genetic diversity in small populations of threatened species {5.3.1} (*speculative*), restoring a proxy of an extinct species {5.3.2} (*speculative*), remediating degraded ecosystems {6.5} (*speculative*), or product replacement {5.2.2, 6.4} (*established but incomplete*).

6. **Synthetic biology and engineered gene drive may be detrimental to conservation and sustainable use of biodiversity {4–6} (*speculative*).** Detrimental effects may stem from the movement of genes, or escape of engineered gene-drive-carrying organisms, impacting non-target populations or species {5.2–5.3, 6.2-6.4} (*speculative*), changes to ecological roles played by target organisms {5.2, 6.3} (*speculative*), broader ecosystem effects {6.2} (*speculative*), product replacement that exacerbates a conservation problem {5.2.2} (*competing explanations*), socio-economic effects of product replacement on livelihoods and on production and consumption patterns {6.4} (*competing explanations*), distracting funding from other conservation approaches {5.1, 5.4} (*speculative*), and moral hazard reducing the urgency and importance of biodiversity conservation {2.3, 5.1} (*speculative*).

7. **Values, worldviews and lived experiences influence the development, assessment and governance of synthetic biology and engineered gene drive {2–3} (*well established*).** Thus, to produce evidence for conservation-relevant decision making, scientific methods and norms operate within contexts defined by the framing of problems and solutions, the integration of multiple perspectives and types of expertise, and who is trusted to produce credible knowledge {3} (*well established*). Community and stakeholder engagement have been proposed to help navigate this complexity {2.3, 3.4} (*established but incomplete*).

8. **Indigenous and local communities are key actors in research, governance and decisions around synthetic biology and engineered gene drive for conservation (*well established*).** Synthetic biology has potentially significant positive and negative impacts on local and indigenous communities, which manage, govern, reside in or depend on a large part of the world's biodiversity {5-6} (*well established*). Historically there has been limited engagement with indigenous and local communities at both the project and global level (*established but incomplete*). Recently there have been calls for recognition of the rights of indigenous and local communities in decision making around synthetic biology and engineered gene drive {2.1} (*well established*). There have been some attempts to involve them in synthetic biology initiatives {2.3} (*established but incomplete*).

9. **Multiple existing governance structures are relevant to synthetic biology (*well established*), but synthetic biology and engineered gene drive raise questions and challenges for these frameworks (*competing explanations*).** Relevant governance frameworks include international, regional and national legal frameworks as well as religious, customary and indigenous governance systems, and scientific norms and practices (*well established*) {2.2}. Challenges relate to the extent to which current and future synthetic biology and gene drive applications are covered by existing regulations, norms and processes (*competing explanations*), implementation and enforcement in the context of accessibility of parts and tools (*established but incomplete*), different levels of governance capacity among jurisdictions (*well established*), mechanisms to address environmental harm, particularly transboundary impacts (*established but incomplete*), and the ability of governance frameworks to keep up with the rapid pace of technological innovation (*competing explanations*) {2.3}.

10. **This "Assessment of Synthetic Biology and Biodiversity Conservation" is neither a risk assessment of individual synthetic biology and gene drive applications, nor of these technologies as a whole {3.4,**

4.3} (*well established*). The diversity of these applications, of the mechanisms that can be used, and of the contexts in which these would take place, precludes an assessment of risks and benefits of this technology as a whole (*well established*). This assessment reviews existing and proposed applications of synthetic biology and engineered gene drive systems that are relevant to conservation and explores how they may be beneficial and detrimental to the conservation and sustainable use of biodiversity. Benefits and risks to conservation from synthetic biology applications vary on a case-by-case basis.

7.2 Looking forward: The IUCN process, interpreting evidence and reaching a policy recommendation

This assessment of synthetic biology and conservation takes places within a broader IUCN conservation policy process. At the 2016 World Conservation Congress in Hawai'i, IUCN's 1,303 government and civil society Members adopted Resolution WCC-2016-Res-086 calling for the establishment of a Task Force to undertake a series of activities to develop an IUCN Synthetic Biology and Biodiversity Conservation Assessment. This assessment will serve as an input to the development of policy recommendations to be debated and voted on by the IUCN membership at the 2020 World Conservation Congress in Marseille.

As directed by the Resolution, all six IUCN Commissions and the Director General appointed the Chair who in turn appointed the Task Force and its Technical Subgroup charged with developing this assessment. This assessment will be finalised based on an open peer review from a DG-appointed expert panel, the entire IUCN constituency and the general public. After revision, the assessment will then feed into policy guidelines that will be drafted by the Task Force and submitted to the IUCN Council (Figure 1.10). Once drafted the policy will receive input from the IUCN Regional Conservation Fora, as well as both online and in-person debate on the motion before being voted on by the full IUCN membership at the 2020 World Conservation Congress. This assessment thus forms one part of IUCN's decision-making process regarding policies to shape the role of synthetic biology in biodiversity conservation. The way IUCN will use the evidence assembled in this assessment to shape the decision will thus be of critical significance.

One primary issue regarding the use of evidence in decision making concerns scientific uncertainty, an underlying issue in environmental governance. As discussed in Chapter 2, under various national and international environmental laws and policies, circumstances in which there is a potential for harm but incomplete or insufficient evidence trigger the precautionary principle [Rio Declaration, Principle 15] (Wiener & Rogers, 2002; Peterson, 2006), which states that where there are threats of serious or irreversible damage, lack of scientific certainty shall not be used as a reason for postponing cost-effective measures to prevent environmental degradation. This in effect places the burden of proof that the action is not unacceptably harmful onto those proposing the action. The precautionary principle is a legal obligation in some countries, and is also an internationally recognised tool for decision making, which may or may not be legally required. In the context of using synthetic biology for conservation the precautionary principle can, however, be utilised to support different positions. These dual interpretations of the precautionary principle are particularly important to surface and discuss given ongoing global biodiversity loss (Butchart et al., 2010) and the insufficiency of existing efforts and methods to prevent it (Maxwell et al., 2016).

The third issue regarding the use of evidence has to do with recognising that subjective judgements and values are always part of any decision-making process, no matter how firmly based in empirical evidence it may be. It is better by far to acknowledge and engage with those values than to only privilege scientific expertise and silence other voices (Chapter 3).

Decision makers must pay attention to factors relating to both the production and use of evidence. Chapter 3 discusses evidence as it relates to assessing the potential impact of synthetic biology on the conservation of biodiversity. The next step

in the IUCN process requires seeing evidence in a slightly different light. This is a challenging topic because of the differing views that exist on what evidence is and how evidence *should* inform decision making. The frameworks, or ways of thinking about, this question of how to incorporate evidence into decision making vary based on the extent to which the framework recognises the interplay of scientific knowledge and public values and to what degree deliberative engagement processes are incorporated. There are three generally recognised frameworks to consider and the third of which is the one closest to the approach taken in creating this assessment.

The *linear* framework envisions the incorporation of evidence into decision making as a technical endeavour, outside of value judgements, that is best completed by experts (Sarewitz, 1996), without any deliberative processes and that scientific experts can accomplish in isolation. The *multiple knowledges* framework acknowledges that there are often tensions between scientific disciplines in terms of how they frame problems and the type of evidence they produce. Without concerted reflection, there is the potential for disciplinary assumptions to delimit both the types of evidence produced and how they are synthesised for a decision-making context.

The *deliberative-analytical* framework highlights the potential for deliberative engagement to be fully incorporated into analytical processes (Sclove, 2010; Delborne et al., 2013; Rask & Worthington, 2015; Bertrand, Pirtle & Tomblin, 2017). One of the foundational assumptions of this model is that analysis and inclusive deliberation are both needed to achieve evidence-based decision making that is both rigorous and legitimate. More analysis and more evidence will not lead to better decision making without the inclusion of values in deliberations informing such analysis and helping to synthesise evidence and make sense of its relevance to a decision-making context. In this approach to incorporating evidence into decision making, scientific experts, decision makers, and interested and affected parties all have a role to play in reviewing evidence and determining its relevance to a decision-making context (NRC, 1996).

This model is best situated for conflictual issues that contain uncertainty – issues that need rigorous deliberative engagement to arrive at an appropriate understanding of the problem, the desired solution, the needed evidence, and how to incorporate existing evidence into a particular decision-making context. In keeping with this approach, IUCN's process is intended to foster rigorous and trusted deliberation across a wide range of experts, affected communities, stakeholders and decision makers in order to successfully develop and deploy a policy on synthetic biology and biodiversity conservation. The review of this assessment, invited from over 15,000 people and organisations located throughout the world is an important facet of this deliberative engagement.

7.3 Technology, society and nature

Conservation and synthetic biology are situated in a landscape that is changing rapidly in at least three dimensions: the technologies underlying synthetic biology are changing at remarkable speed; society is changing in its views about technology and nature, particularly across generations; and nature is changing as well. The three are interlocking: technology changes, society changes in concert, and nature continues to change in response to both. These shifting dynamics provide the broadest context for this assessment.

For decades, the most often cited benchmark for rapid progress in information technology has been Moore's Law, which roughly states that the number of transistors on an integrated circuit doubles every two years (https://www.intel.com/content/www/us/en/silicon-innovations/moores-law-technology.html). That doubling reflects the explosion in computing power that lies at the heart of the revolution in information technology. The technology underlying synthetic biology is now accelerating at an even faster pace. The speed at which scientists can sequence DNA began to outpace Moore's Law in 2008 (Bioeconomy Capital, 2018). Information technology has transformed the way people live and work, and there are good reasons to believe that the changes that will be wrought by synthetic biology will be equally profound and perhaps even more rapid.

Synthetic biology applications are already changing business, industry and medicine. In 2017 the global synthetic biology market was valued at US$ 4.4 billion and is expected to grow to US$ 13.9 billion by 2022 (Globe Newswire, 2018). Private investment appears to be growing rapidly (Figure 1.7). In 2016, investors poured over US$ 1 billion into synthetic biology companies, fuelling their rapid growth. But synthetic biology does not exist in a vacuum; it can interact with nanotechnology, artificial intelligence, robotics and a myriad of biological innovations to yield breakthroughs in smart materials, material structures, energy generation, pollution remediation and more. There is a constant, fluid, and potentially extremely broad interaction and innovation frontier between the Fourth Industrial Revolution and biodiversity.

Scientists are exploring new ways to make changes to the genetic makeup of any species at a speed, specificity and scale unimagined just a few years ago. While the potential future applications appear to be limited only by the imagination, only a relative handful have emerged from laboratory settings (Chapter 5). There remains a great deal of hype for synthetic biology applications and many are speculative or still in early stages of development and testing. Nevertheless, the very existence of the knowledge of how to approach tinkering with the machinery of life raises profound and complex moral, ethical, legal, cultural, spiritual and scientific questions. The breadth and complexity of these questions have resulted in often divergent opinions on the advisability of developing and applying synthetic biology to conservation.

Any new and powerful technology, particularly one with the potential to touch nearly any species and ecological system, anywhere in the world, is a challenge to existing views of what nature is and what should be considered worthy of conservation. Since synthetic biology is still in its early days, most applications have an uncertain future, and societies have not decided if they will support their application. Yet the powerful response to the idea of applying synthetic biology to problems of conservation and sustainable use, both from those wary of the impact and the ethical implication of the new science and those encouraged by the potential of new tools to solve tough challenges, suggests that the impact of synthetic biology on society could be significant.

While synthetic biology may influence society in as yet uncertain ways, the reverse is true as well. As the general public learns more about synthetic biology their opinions will in turn help determine policy decisions about what kind of research receives government funding or regulatory approval, and ultimately which applications will be allowed to take place. Moreover, their demand or preferences as consumers for the products of synthetic biology – or not – will also influence the directions of corporate investment into the field.

Perhaps the most important cultural factor in the future relationship of synthetic biology to conservation will be the attitudes and experiences of young people growing up now with synthetic biology potentially as a fact of life, as well as future generations who will interact with it in ways we cannot predict. Raised in a world in which many technologies are already deployed, younger generations may not share the views of older people to whom these technologies are still novel. There is a generation being raised to consider synthetic biology as just one among many new technologies. For example, the International Genetically Engineered Machine Competition (iGEM) began in 2004 with five teams and 31 participants. In 2018 there were 340 teams, from 42 countries, and 5,806 participants. All told, over 40,000 young people in high school and college – most under the age of 23 – have participated in synthetic biology experiments through iGEM. Many more have been exposed to the field through DIY biology labs now operating around the world or through classroom experience. The application of synthetic biology tools and technologies to conservation will no doubt remain contested, but the attitudes of people now learning about synthetic biology in college or high school biology classes will have an increasingly powerful say in the outcome of the debate.

The ongoing changes in technology and society regarding synthetic biology will presumably lead to changes in nature as well. As with so many of the questions about the intersection of synthetic biology and conservation, the precise contours of those

changes are still uncertain. That there will be changes, however, is beyond debate. Nature itself is changing, and human understanding of nature and natural is changing as well. This has always been the case; the relationship between people and nature has never been static. The pace of that change has accelerated dramatically, however, in tandem with the scale and pace of human transformation of the Earth's biodiversity.

That transformation provides crucial context for assessing the potential impact of synthetic biology on conservation and sustainable use. Not only does the climate, altered by human activity, influence the entire planet, but other human impacts are just as pervasive, from microscopic plastic debris in the farthest reaches of the world's oceans (Galloway, Cole & Lewis, 2017) to persistent organic pollutants in both the Arctic (De March et al., 1998) and Antarctica (Vecchiato et al., 2015). Species extinction caused by people is proceeding a thousand times faster than usual through Earth's history (Pimm et al., 2014).

Technology is an ever more pervasive aspect of the daily lives of people everywhere, including those most remote from urban centres and all the trappings of modernity. This may portend a fundamental shift in the relationship between technology and nature, and some observers argue that to a degree never before seen technology has become an obstacle that prevents humans, particularly children, from experiencing nature as they have through history (Louv, 2008). Just as powerful but less intuitive is the argument that there is no clear distinction between environment and technology, just that technology is – and always has been – the way humans experience the natural world (Reuss & Cutcliffe, 2010). The point here is not to try to resolve the differences between those ways of thinking about the human relationship with nature. The question, which this assessment is designed to help answer, is how diverse communities decide about the conservation uses of technology and whether the environment will benefit or not from these decisions.

The way people will answer that question will depend to a large degree on the way they think about technology, science, society, risk, their perception of their own future and the future of the world around us. Those complex and emotional issues do not exist in isolation, but are tied together by powerful stories that help organise and make sense of the world. As the decision-making processes regarding synthetic biology move forward, the evidence presented in this assessment will become part of new narratives that will help all concerned understand the possibilities and the perils of this new technology.

References, legal instruments and cases

Anon. (2014). *Reply to EFSA's public consultation on Transformation to an Open EFSA*. Available at: https://www.infogm.org/IMG/pdf/transparence_replyfinaldraft_20141001.pdf (Accessed: 25 July 2018).

Abate, W., Sattar, A.A., Liu, J., Conway, M.E. and Jackson S.K. (2017). 'Evaluation of recombinant factor C assay for the detection of divergent lipopolysaccharide structural species and comparison with Limulus amebocyte lysate-based assays and a human monocyte activity assay'. *Journal of Medical Microbiology* 66(7):888–897. https://doi.org/10.1099/jmm.0.000510

Abbadi, A., Domergue, F., Bauer, J., Napier, J.A., Welti, R., Zähringer, U., Cirpus, P. and Heinz, E. (2004). 'Biosynthesis of very-long-chain polyunsaturated fatty acids in transgenic oilseeds: constraints on their accumulation'. *The Plant Cell* 16(10):2734–2748. https://doi.org/10.1105/tpc.104.026070

Abberton, M., Batley, J., Bentley, A., Bryant, J., Cai, H., Cockram, J., Costa de Oliveira, A., Cseke, L.J., Dempewolf, H., De Pace, C., Edwards, D., Gepts, P., Greenland, A., Hall, A.E., Henry, R., Hori, K., Howe, G.T., Hughes, S., Humphreys, M., Lightfoot, D., Marshall, A., Mayes, S., Nguyen, H.T., Ogbonnaya, F.C., Ortiz, R., Paterson, A.H., Tuberosa, R., Valliyodan, B., Varshney, R.K. and Yano, M. (2016). 'Global agricultural intensification during climate change: a role for genomics'. *Plant Biotechnology Journal*. 14(4):1095–1098. https://doi.org/10.1111/pbi.12467

Abbot, C. (2012). 'Bridging the Gap — Non-state Actors and the Challenges of Regulating New Technology'. *Journal of Law and Society* 39(3):329–358. https://doi.org/10.1111/j.1467-6478.2012.00588.x

Abbott, R.C., Osorio, J.E., Bunck, C.M. and Rocke, T.E. (2012). 'Sylvatic plague vaccine: a new tool for conservation of threatened and endangered species?'. *EcoHealth* 9(3):243–250. https://doi.org/10.1007/s10393-012-0783-5

Ackerman, F. and Heinzerling, L. (2004). *Priceless: On Knowing the Price of Everything and the Value of Nothing*.

Ad Hoc Technical Expert Group (AHTEG) on Digital Sequence Information on Genetic Resources (2018a). *Report of the AHTEG on Digital Sequence Information*. Available at: https://www.cbd.int/doc/c/4f53/a660/20273cadac313787b058a7b6/dsi-ahteg-2018-01-04-en.pdf.

Ad Hoc Technical Expert Group (AHTEG) on Digital Sequence Information on Genetic Resources (2018b). *Synthesis of Views and Information on the Potential Implications of the Use of Digital Sequence Information on Genetic Resources for the Three Objectives of the Convention and the Objective of the Nagoya Protocol. CBD/DSI/AHTEG/2018/1/4*. Montreal. Available at: https://www.cbd.int/doc/c/49c9/06a7/0127fe7bc6f3bc5a8073a286/dsi-ahteg-2018-01-02-en.pdf.

Ad Hoc Technical Expert Group on Synthetic Biology (2015). *Report of the Ad Hoc Technical Expert Group on Synthetic Biology*. Montreal. Available at: http://bch.cbd.int/synbio/open- (Accessed: 1 August 2018).

Ad Hoc Technical Expert Group on Synthetic Biology (2017). *Report of the Ad Hoc Technical Expert Group on Synthetic Biology*. Montreal.

Ad Hoc Technical Expert Group on Synthetic Biology (2018). *Report of the Ad Hoc Technical Expert Group on Synthetic Biology*. Montreal.

Adams, W. M. (2017). 'Geographies of conservation I: De-extinction and precision conservation'. *Progress in Human Geography* 41(4):534–545. https://doi.org/10.1177/0309132516646641

Adarme-Vega, T.C., Lim, D.K.Y., Timmins, M., Vernen, F., Li, Y. and Schenk, P.M. (2012). 'Microalgal biofactories: a promising approach towards sustainable omega-3 fatty acid production'. *Microbial cell factories* 11(1):96. https://doi.org/10.1186/1475-2859-11-96

Africa Geographic (2015). *Biologist aims to grow synthetic rhino horns*, *Africa Geographic*. Available at: https://africageographic.com/blog/biologist-aims-to-grow-synthetic-rhino-horns/

African Union (2014). 'African Union Executive Council Twenty-Fourth Ordinary Session Addis Ababa Ethiopia', 812(January), pp. 25–26.

African Union (2018). *Gene drives for malaria control and elimination in Africa*.

Akbari, O. S., Bellen, H.J., Bier, E., Bullock, S. L., Burt, A., Church, G.M., Cook, K.R., Duchek, P., Edwards, O.R., Esvelt, K.M., Gantz, V.M., Golic, K.G., Gratz, S.J., Harrison, M.M., Hayes, K.R., James, A.A., Kaufman, T.C., Knobich, J., Malik, H.S., Matthews, K.A., O'Connor-Giles, K.M., Parks, A.L., Perrimon, N., Port, F., Russell, S., Ueda, R. and Wildonger, J. (2015). 'Safeguarding

gene drive experiments in the laboratory'. *Science* 349(6251):927–929. https://doi.org/10.1126/science.aac7932

Akin, H., Rose, K.M., Scheufele, D.A., Simis-Wilkinson, M., Brossard, D., Xenos, M.A. and Corley, E.A. (2017). 'Mapping the landscape of public attitudes on synthetic biology'. *BioScience* 67(3):290–300. https://doi.org/10.1093/biosci/biw171

Alexander, P., Brown, C., Arneth, A., Dias, C., Finnigan, J., Moran, D. and Rounsevella, M.D.A. (2017). 'Could consumption of insects, cultured meat or imitation meat reduce global agricultural land use?' *Global Food Security* 15:22–32. https://doi.org/10.1016/j.gfs.2017.04.001

Alexandratos, N. and Bruinsma, J. (2012). *World agriculture towards 2030/2050: the 2012 revision*. ESA Working paper FAO, Rome.

Amalric, F. (2005). 'The equator principles'. *Finance & Bien Commun* (2):8–11. https://doi.org/10.3917/fbc.022.0008

Amante-Helweg, V.L.U. and Conant, S. (2009). 'Hawaiian culture and forest birds'. In: T.K. Pratt, C.T. Atkinson, P.C. Banko, J.D. Jacobi and B.L. Woodworth (eds.) *Conservation Biology of Hawaiian Forest Birds: Implications for Island Avifauna*, pp.59–82. New Haven: Yale University Press.

Amyris (2018). *Clean Beauty*. Available at: https://amyris.com/product-category/clean-beauty/ (Accessed: 16 July 2018).

Anagnostakis, S. L. (1987). 'Chestnut blight: the classical problem of an introduced pathogen'. *Mycologia* 79(1):23–37. https://doi.org/10.1080/00275514.1987.12025367

Andersson, J. O. (2005). 'Lateral gene transfer in eukaryotes'. *Cellular and Molecular Life Sciences* 62:1182–1197. https://doi.org/10.1007/s00018-005-4539-z

Antolin, M.F., Gober, P., Luce, B., Biggins, D.E., Van Pelt, W.E., Seery, D.B., Lockhart, M. and Ball, M. (2002). 'The influence of sylvatic plague on North American wildlife at the landscape level, with special emphasis on black-footed ferret and prairie dog conservation'. US Fish & Wildlife Publications.

Antúnez, K., Antúnez, K., Martín-Hernández, R., Prieto, L., Meana, A., Zunino, P. and Higes, M. (2009). 'Immune suppression in the honey bee (*Apis mellifera*) following infection by *Nosema ceranae* (*Microsporidia*)'. *Environmental Microbiology* 11(9):2284–2290. https://doi.org/10.1111/j.1462-2920.2009.01953.x.

Arendt, K.E., Jónasdóttir, S.H., Hansen, P.J. and Gärtner, S. (2005). 'Effects of dietary fatty acids on the reproductive success of the calanoid copepod Temora longicornis'. *Marine Biology* 146(3):513–530. https://doi.org/10.1007/s00227-004-1457-9

Arita, I. (1980). 'Smallpox Eradication: Man's Success in Eliminating a Most Dangerous Disease'. *Environmental Conservation* 7(03):176. https://doi.org/10.1017/S0376892900007487.

Arrowsmith, J. (2011). 'Trial watch: Phase II failures: 2008–2010'. Nature Publishing Group.

Ascher, W., Steelman, T.A. and Healy, R.G. (2010). *Knowledge and environmental policy: Re-imagining the boundaries of science and politics*. MIT Press Cambridge, MA. https://doi.org/10.7551/mitpress/8398.001.0001

Atkinson, C.T., Woods, K.L., Dusek, R.J., Sileo, L.S. and Iko, W.M. (1995). 'Wildlife disease and conservation in Hawaii: pathogenicity of avian malaria (*Plasmodium relictum*) in experimentally infected Iiwi (*Vestiaria coccinea*)'. *Parasitology* 111(S1):S59–S69. https://doi.org/10.1017/S003118200007582X

Atkinson, C.T., Saili, K.S., Utzurrum, R.B. and Jarvi, S.I. (2013). 'Experimental evidence for evolved tolerance to avian malaria in a wild population of low elevation Hawai'i' Amakihi (*Hemignathus virens*)'. *EcoHealth* 10(4):366–375. https://www.ncbi.nlm.nih.gov/pubmed/24430825

Atkinson, C.T., Utzurrum, R.B., Lapointe, D.A., Camp, R.J., Crampton, L.H., Foster, J.T. and Giambelluca, T.W. (2014). 'Changing climate and the altitudinal range of avian malaria in the Hawaiian Islands–an ongoing conservation crisis on the island of Kaua'i'. *Global Change Biology* 20(8):2426–2436. https://doi.org/10.1111/gcb.12535

Atkinson, C.T. and LaPointe, D.A. (2009a). 'Ecology and pathogenicity of avian malaria and pox'. In: T.K. Pratt, C.T. Atkinson, P.C. Banko, J.D. Jacobi and B.L. Woodworth (eds.) *Conservation biology of Hawaiian forest birds: Implications for island avifauna*, p.252. Yale University Press New York.

Atkinson, C.T. and LaPointe, D.A. (2009b). 'Introduced avian diseases, climate change, and the future of Hawaiian honeycreepers'. *Journal of Avian Medicine and Surgery* 23(1):53–63.

Atkinson, I. A. E. (1985). 'The spread of commensal species of Rattus to oceanic islands and their effects on island avifaunas'. In: P. J. Moors (ed.) *Conservation of Island Birds*, pp. 35–81. London: ICBP Technical Publication No. 3.

Atlantic States Marine Fisheries Commission (2015). *Atlantic States Marine Fisheries Commission: A Framework for Adaptive Management of Horseshoe Crab*, Report.

Atyame, C.M., Cattel, J., Lebon, C., Flores, O., Dehecq, J., Weill, M., Gouagna, L.C. and Tortosa, P. (2015). 'Wolbachia-based population control strategy targeting *Culex quinquefasciatus* mosquitoes proves efficient under semi-field conditions'. *PloS one* 10(3): e0119288. https://doi.org/10.1371/journal.pone.0119288

Atyame, C. M., Labbé, P., Lebon, C., Weill, M., Moretti, R., Marini, F., Gouagna, L.C., Calvitti, M. and Tortosa, P. (2016). 'Comparison of irradiation and Wolbachia based approaches for sterile-male strategies targeting *Aedes albopictus*'. *PLoS one* 11(1): e0146834. https://doi.org/10.1371/journal.pone.0146834

August, P. R., Grossman, T.H., Minor, C., Draper, M.P., MacNeil, I.A., Pemberton, J.M., Call, K.M., Holt, D., Osburne, M.S. (2000). 'Sequence analysis and functional characterization of the violacein biosynthetic pathway from *Chromobacterium violaceum*'. *Journal of molecular microbiology and biotechnology* 2(4):513–519.

Austin, H.P., Austin, H.P., Allen, M.D., Donohoe, B.S., Rorrer, N.A., Kearns, F.L., Silveira, R.L., Pollard, B.C., Dominick, G., Duman, R., Omari, K.E., Mykhaylyk, V., Wagner, A., Michener, W.E., Amore, A., Skaf, M.S., Crowley, M.F., Thorne, A.W., Johnson, C.W., Woodcock, H.L., McGeehan, J.E. and Beckham, G.T. (2018). 'Characterization and engineering of a plastic-degrading aromatic polyesterase'. *Proceedings of the National Academy of Sciences* 115(19):E4350–E4357. https://doi.org/10.1073/pnas.1718804115

Australian Academy of Sciences (AAS) (2017). *Synthetic Gene Drives in Australia: Implications of Emerging Technologies*. Canberra. Available at: https://www.science.org.au/support/analysis/reports/synthetic-gene-drives-australia-implications-emerging-technologies

Ba, H., Jia, B., Wang, G., Yang, Y., Kedem, G. and Li, C. (2017). 'Genome-Wide SNP Discovery and Analysis of Genetic Diversity in Farmed Sika Deer (*Cervus nippon*) in Northeast China Using Double-Digest Restriction-Site Associated DNA Sequencing'. *G3: Genes, Genomes, Genetics*. G3: Genes, Genomes, Genetics, https://doi.org/10.1534/g3.117.300082

Babcock, R.C. and Mundy, C.N. (1992). 'Reproductive biology, spawning and field fertilization rates of Acanthatser planci'. *Marine and Freshwater Research*. CSIRO, 43(3), pp. 525–533. https://doi.org/10.1071/MF9920525

Bäckstrand, K., Khan, J., Kronsell, A. and Lovbrand, E. (2010). 'The promise of new modes of environmental governance'. In: *Environmental Politics and Deliberative Democracy*. Edward Elgar Publishing.

Bagley, M. A. (2016). *Digital DNA: The Nagoya Protocol, Intellectual Property Treaties, and Synthetic Biology*.

Bagley, M. A. and Rai, A. (2013). *The Nagoya Protocol and Synthetic Biology Research: A Look at the Potential Impacts*. Washington, D.C.

Bagley, M. and Rai, A. (2014). 'The Nagoya Protocol and Synthetic Biology Research: a look at the potential impacts'.

Baker, D.M., Freeman, C. J., Wong, J.C.Y., Fogel, M.L. and Knowlton, N. (2018). 'Climate change promotes parasitism in a coral symbiosis'. *The ISME journal* 12(3):921.

Baker, M. (2016). 'Statisticians issue warning on P values'. *Nature* 531(7593):151.

Balakrishna, P., Dharmaji, B. and Warner, E. (2003). 'Risk assessment and risk management in implementing the Cartagena protocol: proceedings of Asia regional workshop'. https://portals.iucn.org/library/node/8326

Balmer, A. and Martin, P. (2008). 'Synthetic biology. Social and ethical challenges. An independent review commissioned by the Biotechnology and Biological Sciences Research Council (BBSRC)'. *Institute for Science and Society, University of Nottingham*, pp. 1–36. Available at: www.bbsrc.ac.uk/web/files/reviews/0806_synthetic_biology.pdf

Banach, M., Edholm, E. and Robert, J. (2017). 'Exploring the functions of nonclassical MHC class Ib genes in *Xenopus laevis* by the CRISPR/Cas9 system'. *Developmental Biology* 426(2):261–269. https://doi.org/10.1016/j.ydbio.2016.05.023

Bardwell, L.V. (1991). 'Problem-framing: a perspective on environmental problem-solving'. *Environmental Management* 15(5):603–612. https://doi.org/10.1007/BF02589620

Barichievy, C., Sheldon, R. Wacher, T., Llewellyn, O., Al-Mutairy, M., Alagaili, A. (2018). 'Conservation in Saudi Arabia; moving from strategy to practice'. *Saudi Journal of Biological Sciences* 25(2):290–292. https://doi.org/10.1016/j.sjbs.2017.03.009

Bateman, R., Sulaiman, J.M. and Ginting, T.J. (2014). *ASEAN Guidelines on the Regulation, Use, and Trade of Biological Control Agents (BCA).*

Bates, K.A. Clare, F.C., O'Hanlon, S., Bosch, J., Brookes, L., Hopkins, K., McLaughlin, E.J., Daniel, O., Garner, T.W.J., Fisher, M. C. and Harrison, X. A. (2018). 'Amphibian chytridiomycosis outbreak dynamics are linked with host skin bacterial community structure'. *Nature Communications* 9(1):693. https://doi.org/10.1038/s41467-018-02967-w

Becker, M.H., Harris, R.N., Minbiole, K.P.C., Schwantes, C.R., Rollins-Smith, L.A., Reinert, L.K., Brucker, R.M., Domangue, R.J. and Gratwicke, B. (2011). 'Towards a better understanding of the use of probiotics for preventing chytridiomycosis in Panamanian golden frogs'. *Ecohealth* 8(4):501–506. https://doi.org/10.1007/s10393-012-0743-0

Becker, S., Bryman, A. and Ferguson, H. (2012). *Understanding research for social policy and social work: themes, methods and approaches*. Policy Press. https://doi.org/10.2307/j.ctt1t892hf

Begley, C.G. and Ellis, L.M. (2012). 'Drug development: Raise standards for preclinical cancer research'. *Nature* 483(7391):531. https://doi.org/10.1038/483531a

Bellard, C., Cassey, P. and Blackburn, T.M. (2016). 'Alien species as a driver of recent extinctions'. *Biology Letters* 12(2):20150623. https://doi.org/10.1098/rsbl.2015.0623

Bellard, C., Genovesi, P. and Jeschke, J.M. (2016). 'Global patterns in threats to vertebrates by biological invasions'. *Proceedings of the Royal Society of London B: Biological Sciences* 283(1823). https://doi.org/10.1098/rspb.2015.2454

Bender, S.F., Wagg, C. and van der Heijden, M.G.A. (2016). 'An underground revolution: biodiversity and soil ecological engineering for agricultural sustainability'. *Trends in Ecology & Evolution* 31(6):440–452. https://doi.org/10.1016/j.tree.2016.02.016

Bennett, E.L. (2015). 'Legal ivory trade in a corrupt world and its impact on African elephant populations'. *Conservation Biology* 29(1):54–60. https://doi.org/10.1111/cobi.12377

Benton, M.J. (2015). *When life nearly died: the greatest mass extinction of all time*. Revised Ed. Thames & Hudson.

Bergel, S.D. (2015). 'Patentability of human genes: The conceptual differences between the industrialised and Latin American countries'. *Journal of Community Genetics* 6(3):321–327. https://doi.org/10.1007/s12687-015-0228-2

Bergeson, L., Dolan, S.L. and Engler, R.E. (2015). 'The DNA of the US Regulatory System: Are We Getting It Right for Synthetic Biology?' *Woodrow Wilson Center Project on Synthetic Biology* 41–43.

Bernardini, F., Galizi, R., Wunderlich, M., Taxiarchi, C., Kranjc, N., Kyrou, K., Hammond, A., Nolan, T., Lawniczak, M.N.K., Papathanos, P.A., Crisanti, A. and Windbichler, N. (2017). 'Cross-species Y chromosome function between malaria vectors of the Anopheles gambiae species complex'. *Genetics*. https://doi.org/10.1534/genetics.117.300221

Bertrand, P., Pirtle, Z. and Tomblin, D. (2017). 'Participatory technology assessment for Mars mission planning: Public values and rationales'. *Space Policy* 42:41–53. https://doi.org/10.1016/j.spacepol.2017.08.004

Best, M.L. and Hartwell, H.J. (2014). 'The trophic role of a forest salamander: impacts on invertebrates, leaf litter retention, and the humification process'. *Ecosphere* 5(2):1–19. https://doi.org/10.1890/ES13-00302.1

Bhatt, S. Weiss, D.J., Cameron, E., Bisanzio, D., Mappin, B., Dalrymple, U., Battle, K. E., Moyes, C. L., Henry, A., Eckhoff, P.A., Wenger, E.A., Briët, O., Penny, M.A., Smith, T. A., Bennett, A., Yukich, J., Eisele, T.P., Griffin, J.T., Fergus, C.A., Lynch, M., Lindgren, F., Cohen, J.M., Murray, C.L.J., Smith, D.L., Hay, S.I., Cibulskis, R.E. and Gething, P.W. (2015). 'Europe PMC Funders Group The effect of malaria control on Plasmodium falciparum in Africa between 2000 and 2015'. *Nature, available in PMC*, 526(7572):207–211. https://doi.org/10.1038/nature15535

Biggins, D.E. and Schroeder, M.H. (1988). 'Historical and present status of the black-footed ferret'. In: *Eighth Great Plains Wildlife Control Workshop, Rapid City, South Dakota*.

Bilotta, G.S., Milner, A.M. and Boyd, I. (2014). 'On the use of systematic reviews to inform environmental policies'. *Environmental Science & Policy* 42:67–77. https://doi.org/10.1016/j.envsci.2014.05.010

Biodesign Challenge (2018). *Biodesign Challenge*. Available at: http://biodesignchallenge.org/ (Accessed: 16 July 2018).

Bioeconomy Capital (2018). *Bioeconomy Dashboard*. Available at: http://www.bioeconomycapital.com/bioeconomy-dashboard/ (Accessed: 9 November 2018).

Biotechnology and Biological Sciences Research Council of the United Kingdom (BBSRC UK) (2017). *Capacity building for the bioeconomy in Africa*.

Birnie, P., Boyle, A. and Redgwell, C. (2009). *International Law and the Environment*. Third Edit. Oxford University Press.

Blackall, L.L., Wilson, B. and van Oppen, M.J.H. (2015). 'Coral—the world's most diverse symbiotic ecosystem'. *Molecular Ecology* 24(21):5330–5347. https://doi.org/10.1111/mec.13400

Blake, S., Deem, S.L., Mossimbo, E., Maisels, F., Walsh, P. (2009). 'Forest elephants: tree planters of the Congo'. *Biotropica* 41(4):459–468. https://doi.org/10.1111/j.1744-7429.2009.00512.x

Bland, L.M., Keith, D.A., Miller, R.M., Murray, N.J. and Rodriguez, J.P. (2015). *Guidelines for the application of IUCN Red List of Ecosystems Categories and Criteria, Version 1.0*. Gland, Switzerland: IUCN. https://doi.org/10.2305/IUCN.CH.2016.RLE.1.en

Blehert, D.S., Hicks, A.C., Behr, M., Meteyer, C.U., Berlowski-Zier, B.M., Buckles, E.L., Coleman, J.T.H., Darling, S.R., Gargas, A., Niver, R., Okoniewski, J.C., Rudd, R.J. and Stone, W.B. (2009). 'Bat white-nose syndrome: an emerging fungal pathogen?'. *Science* 323(5911):227. https://doi.org/10.1126/science.1163874

Bletz, M.C., Loudon, A.H., Becker, M.H., Bell, S.C., Woodhams, D.C., Minbiole, K.P.C. and Harris, R.N. (2013). 'Mitigating amphibian chytridiomycosis with bioaugmentation: characteristics of effective probiotics and strategies for their selection and use'. *Ecology Letters* 16(6):807–820. https://doi.org/10.1111/ele.12099

Bloomberg (2018). 'This Biohacker Makes Mutant Frogs — And You Can Buy Them on the Internet'. *Fortune* September. Available at: http://fortune.com/2018/09/12/genetic-engineering-buy-frogs/

Bodmer, R., Pezo Lozano, E. and Fang, T. (2004). 'Economic analysis of wildlife use in the Peruvian Amazon'. In: K. Silvius, R. Bodmer and J. Fragoso. (eds.) *People in Nature: wildlife conservation in South and Central America*, pp. 191–207. New York: Columbia University Press. https://doi.org/10.7312/silv12782-012

Bolden, J. (2018). 'Personal Communications'.

Bolden, J. and Mozier, N. (2018). 'Personal Communications'.

Bomford, M. and O'Brien, P. (1995). 'Eradication or control for vertebrate pests?'. *Wildlife Society Bulletin (1973-2006).* 23(2):249–255.

Bomgardner, M.M. (2016). 'The Problem with Vanilla'. *Chemical & Engineering News*, September.

Bono, J.M., Olesnicky, E.C. and Matzkin, L.M. (2015). 'Connecting genotypes, phenotypes and fitness: harnessing the power of CRISPR/Cas9 genome editing'. *Molecular Ecology* 24(15):3810–3822. https://doi.org/10.1111/mec.13252

Borel, B. (2017). 'CRISPR, microbes and more are joining the war against crop killers'. *Nature* 543(7645):302–304. https://doi.org/10.1038/543302a.

Bourzac, K. (2017). 'Bioengineering: Solar upgrade'. *Nature* 544(7651): S11–S13. https://doi.org/10.1038/544S11a

Bromenshenk, J.J., Henderson, C.B., Wick, C.H., Stanford, M.F., Zulich, A. W., Jabbour, R.E., Deshpande, S.V., McCubbin, P.E., Seccomb, R.A., Welch, P.M., Williams, T., Firth, D.R., Skowronski, E., Lehmann, M.M., Bilimoria, S.L., Gress, J., Wanner, K.W. and Cramer Jr, R.A. (2010). 'Iridovirus and microsporidian linked to honey bee colony decline'. *PloS one* 5(10):e13181. https://doi.org/10.1371/journal.pone.0013181

Broome, K., Cox, A., Golding, C., Cromarty, P., Bell, P. and McClelland, P. (2014). 'Rat eradication using aerial baiting Current agreed best practice used in New Zealand (Version 3.0)'. New Zealand Department of Conservation (ed.). Wellington, New Zealand.: New Zealand Department of Conservation.

Brown Weiss, E. (1993). 'Intergenerational equity: toward an international legal framework'. In: E. B. Weiss (ed.) *Environmental change and international law: New challenges and dimensions*, pp. 333–354. Tokyo: United Nations Press.

Brune, K.D. and Bayer, T. (2012). 'Engineering microbial consortia to enhance biomining and bioremediation'. *Frontiers in Microbiology* 3:203. https://doi.org/10.3389/fmicb.2012.00203

Bruno, J.F., Bates, A.E., Cacciapaglia, C., Pike, E. P., Amstrup, S. C., van Hooidonk, R., Henson, S. A., Aronson and R. B. (2018). 'Climate change threatens the world's marine protected areas'. *Nature Climate Change*. https://doi.org/10.1038/s41558-018-0149-2

Burkard, C., Lillico, S.G., Reid, E., Jackson, B., Mileham, A.J., Ait-Ali, T., Whitelaw, C.B.A. and Archibald, A.L. (2017). 'Precision engineering for PRRSV resistance in pigs: macrophages from genome edited pigs lacking CD163 SRCR5 domain are fully resistant to both PRRSV genotypes while maintaining biological function'. *PLoS Pathogens*13(2):e1006206. https://doi.org/10.1371/journal.ppat.1006206

Burt, A. (2003). 'Site-specific selfish genes as tools for the control and genetic engineering of natural populations'. *Proceedings of the Royal Society of London B: Biological Sciences* 270(1518):921–928. https://doi.org/10.1098/rspb.2002.2319

Burt, A., Coulibaly, M., Crisanti, A., Diabate, A. and Kayondo, J. (2018). 'Gene drive to reduce malaria transmission in sub-Saharan Africa'. *Journal of Responsible Innovation* 5:S66–S80. https://doi.org/10.1080/23299460.2017.1419410

Burt, A. and Crisanti, A. (2018). 'Gene Drive: Evolved and Synthetic'. *ACS Chemical Biology* 13(2):343–346. https://doi.org/10.1021/acschembio.7b01031

Burt, A. and Trivers, R. (2006). *Genes in conflict: the biology of selfish genetic elements*. Belknap Press of Harvard University Press. https://doi.org/10.4159/9780674029118

Burton, A. (2009). 'PESTICIDES: toward DDT-free malaria control.' *Environmental Health Perspectives* 117(8). https://doi.org/10.1289/ehp.117-a344

Burton, T. M. and Likens, G. E. (1975). 'Salamander populations and biomass in the Hubbard Brook experimental forest, New Hampshire'. *Copeia* 1975(3):541–546. https://doi.org/10.2307/1443655

Butchart, S.H.M., Walpole, M., Collen, B., van Strien, A., Scharlemann, J.P.W., Almond, R.E.A., Baillie, J.E.M., Bomhard, B., Brown, C., Bruno, J., Carpenter, K.E., Carr, G.M., Chanson, J., Chenery, A.M., Csirke, J., Davidson, N.C., Dentener, F., Foster, M., Galli, A., Galloway, J.N., Genovesi, P., Gregory, R.D., Hockings, M., Kapos, V., Lamarque, J., Leverington, F., Loh, J., McGeoch, M.A., McRae, L., Minasyan, A., Morcillo, M.H., Oldfield, T.E.E., Pauly, D., Quader, S., Revenga, C., Sauer, J.R., Skolnik, B., Spear, D., Stanwell-Smith, D., Stuart, S.N., Symes, A., Tierney, M., Tyrrell, T.D., Vié, J. and Watson, R. (2010). 'Global biodiversity: indicators of recent declines'. *Science* 328(5982):1164–1168. https://doi.org/10.1126/science.1187512

Butchart, S.H.M., Scharlemann, J.P.W., Evans, M.I., Quader, S., Aricò, S., Arinaitwe, J., Balman, M., Bennun, L.A., Bertzky, B., Besançon, C., Boucher, T.M., Brooks, T.M., Burfield, I.J., Burgess, N.D., Chan, S., Clay, R.P., Crosby, M.J., Davidson, N.C., De Silva, N., Devenish, C., Dutson, G.C.L., Díaz Fernández, D.F., Fishpool, L.D.C., Fitzgerald, C., Foster, M., Heath, M.F., Hockings, M., Hoffmann, M., Knox, D., Larsen, F.W., Lamoreux, J.F., Loucks, C., May, I., Millett, J., Molloy, D., Morling, P., Parr, M., Ricketts, T.H., Seddon, N., Skolnik, B., Stuart, S.N., Upgren, A. and Woodley, S. (2012). 'Protecting important sites for biodiversity contributes to meeting global conservation targets'. *PloS one* 7(3):e32529. https://doi.org/10.1371/journal.pone.0032529

Callebaut, S. (2015). *New developments in modern biotechnology: A survey and analysis of the regulatory status of plants produced through New Breeding techniques*. Ghent University. Available at: https://lib.ugent.be/fulltxt/RUG01/002/213/647/RUG01-002213647_2015_0001_AC.pdf

Calvert, J. (2008). 'The commodification of emergence: systems biology, synthetic biology and intellectual property'. *BioSocieties* 3(4):383–398. https://doi.org/10.1017/S1745855208006303

Calvert, J. (2012). 'Ownership and sharing in synthetic biology: A "diverse ecology" of the open and the proprietary?'. *BioSocieties* 7(2):169–187. https://doi.org/10.1057/biosoc.2012.3

Campbell, B.M., Beare, D.J., Bennett, E.M., Hall-Spencer, J.M., Ingram, J.S.I., Jaramillo, F., Ortiz, R., Ramankutty, N., Sayer, J.A. and Shindell, D. (2017). 'Agriculture production as a major driver of the Earth system exceeding planetary boundaries'. *Ecology and Society* 22(4):8. https://doi.org/10.5751/ES-09595-220408

Campbell, K. and Donlan, C.J. (2005). 'Feral goat eradications on islands'. *Conservation Biology* 19(5):1362–1374. https://doi.org/10.1111/j.1523-1739.2005.00228.x

Campbell, K.J., Beek, J., Eason, C.T., Glen, A.S., Godwin, J., Gould, F., Holmes, N.D., Howald, G.R., Madden, F.M., Ponder, J.B., Threadgill D.W., Wegmann, A.S. and Baxter, G.S. (2015). 'The next generation of rodent eradications: innovative technologies and tools to improve species specificity and increase their feasibility on islands'. *Biological Conservation* 185:47–58. https://doi.org/10.1016/j.biocon.2014.10.016

Carlson, R. (2005). 'Splice It Yourself'. *Wired*, May.

Carman, T. (2018). 'A new way to fish'. *The Washington Post*, 13 November. Available at: https://www.washingtonpost.com/graphics/2018/lifestyle/cultured-bluefin-tuna/?utm_term=.79758d0e50e2

Carmichael, R.H., Botton, M.L., Shin, P.K.S. and Cheung, S.G. (eds.) (2015). *Changing Global Perspectives on Horseshoe Crab Biology, Conservation and Management*. Springer. https://doi.org/10.1007/978-3-319-19542-1

Casadesús, J. and Low, D. (2006). 'Epigenetic gene regulation in the bacterial world'. *Microbiology and Molecular Biology Reviews* 70(3):830–856.

CBD/SBSTTA/22/CRP.10 (2018). *Digital sequence information on genetic resources*. *Note by the Executive Secretary*. Available at: https://www.cbd.int/abs/dsi-gr/ahteg.shtml#peerreview. (Accessed: 1 August 2018).

Ceballos, G. and Ehrlich, P.R. (2018). 'The misunderstood sixth mass extinction'. *Science* 360(6393):1080–1081. https://doi.org/10.1126/science.aau0191

Ceballos, G., Ehrlich, P.R. and Dirzo, R. (2017). 'Biological annihilation via the ongoing sixth mass extinction signaled by vertebrate population losses and declines'. *Proceedings of the National Academy of Sciences of the United States of America* 114(30):E6089–E6096. https://doi.org/10.1073/pnas.1704949114

Chakravarti, L.J., Beltran, V.H. and van Oppen, M.J.H. (2017). 'Rapid thermal adaptation in photosymbionts of reef-building corals'. *Global Change Biology* 23(11):4675–4688. https://doi.org/10.1111/gcb.13702

Challender, D.W.S., Harrop, S.R. and MacMillan, D.C. (2015). 'Towards informed and multi-faceted wildlife trade interventions'. *Global Ecology and Conservation* 3:129–148. https://doi.org/10.1016/j.gecco.2014.11.010

Champer, J., Liu, J., Oh, S.Y., Reeves, R., Luthra, A., Oakes, N., Clark, A.G. and Messer, P.W. (2018). 'Reducing resistance allele formation in CRISPR gene drive'. *Proceedings of the National Academy of Sciences* 115(21): 5522–5527. https://doi.org/10.1073/pnas.1720354115

Champer, J., Buchman, A. and Akbari, O.S. (2016). 'Cheating evolution: engineering gene drives to manipulate the fate of wild populations'. *Nature Reviews Genetics* 17(3):146. https://doi.org/10.1038/nrg.2015.34

Chan, W.Y., Peplow, L.M., Menéndez, P., Hoffmann, A.A. and van Oppen, M.J.H. (2018). 'Interspecific Hybridization May Provide Novel Opportunities for Coral Reef Restoration'. *Frontiers in Marine Science* 5:160. https://doi.org/10.3389/fmars.2018.00160

Charles, D. (2001). *Lords of the harvest: Biotech, big money, and the future of food*. Perseus Books Group.

Charo, R.A. and Greely, H.T. (2015). 'CRISPR critters and CRISPR cracks'. *The American Journal of Bioethics* 15(12):11–17. https://doi.org/10.1080/15265161.2015.1104138

Chaudhary, A., Pfister, S. and Hellweg, S. (2016). 'Spatially explicit analysis of biodiversity loss due to global agriculture, pasture and forest land use from a producer and consumer perspective'. *Environmental Science & Technology* 50(7):3928–3936. https://doi.org/10.1021/acs.est.5b06153

Chen, C., Sun, Q., Narayanan, B., Nuss, D.L. and Herzberg, O. (2010). 'Structure of oxaloacetate acetylhydrolase, a virulence factor of the chestnut blight fungus'. *Journal of Biological Chemistry* 285:26685–26696. https://doi.org/10.1074/jbc.M110.117804

Cheng, T.L., Mayberry, H., McGuire, L.P., Hoyt, J.R., Langwig, K.E., Nguyen, H., Parise, K.L., Foster, J.T., Willis, C.K.R., Kilpatrick, A.M. and Frick, W.F. (2017). 'Efficacy of a probiotic bacterium to treat bats affected by the disease white-nose syndrome'. *Journal of Applied Ecology* 54(3):701–708. https://doi.org/10.1111/1365-2664.12757

Commonwealth Head of Government Meeting (CHOGM) (2018). *Malaria summit commitments*.

Church of Scotland (2010). 'Synthetic Biology'. Church and Society Council.

Clavero, M. and García-Berthou, E. (2005). 'Invasive species are a leading cause of animal extinctions'. *Trends in Ecology & Evolution* 20(3):110. https://doi.org/10.1016/j.tree.2005.01.003

Cleves, P.A., Strader, M.E., Bay, L.K., Pringle, J.R. and Matz, M.V. (2018). 'CRISPR/Cas9-mediated genome editing in a reef-building coral'. *Proceedings of the National Academy of Sciences* 115(20):5235–5240. https://doi.org/10.1073/pnas.1722151115

Clulow, J., Trudeau, V.L. and Kouba, A.J. (2014). 'Amphibian declines in the twenty-first century: why we need assisted reproductive technologies'. In: *Reproductive Sciences in Animal Conservation*, pp. 275–316. Springer. https://doi.org/10.1007/978-1-4939-0820-2_12

Collins, C.M., Bonds, J.A.S., Quinlan, M.M. and Mumford, J.D. (2018). 'Effects of the removal or reduction in density of the malaria mosquito, *Anopheles gambiae s.l.*, on interacting predators and competitors in local ecosystems'. *Medical and Veterinary Entomology*. https://doi.org/10.1111/mve.12327

Collins, H.M. (1983). 'An empirical relativist programme in the sociology of scientific knowledge'. In: K. Knorr-Cetina and M. Mulkay (eds.) *Science Observed*. London: Sage.

Colombo, S.M., Campbell, L.G., Murphy, E.J., Martin, S.L. and Arts, M.T. (2018). 'Potential for novel production of omega-3 long-chain fatty acids by genetically engineered oilseed plants to alter terrestrial ecosystem dynamics'. *Agricultural Systems* 164:31–37. https://doi.org/10.1016/j.agsy.2018.03.004

Coluzzi, M., Sabatini, A., Petrarca, V. and Di Deco, M.A. (1979). 'Chromosomal differentiation and adaptation to human environments in the *Anopheles gambiae* complex'. *Transactions of the Royal Society of Tropical Medicine and Hygiene*. https://doi.

org/10.1016/0035-9203(79)90036-1

Commission of the Episcopates of the European Union (COMECE) (2016). *Opinion of the Reflection Group on Bioethics on Synthetic Biology*. Brussels, Belgium: COMECE.

Conlon, J.M. (2011). 'Structural diversity and species distribution of host-defense peptides in frog skin secretions'. *Cellular and Molecular Life Sciences* 68(13):2303–2315. https://doi.org/10.1007/s00018-011-0720-8

Conner, N. (2016). *Socio-economic dimensions of human dependence on nature. A review of conceptual frameworks, tools and methodologies used in assessment*.

Conservation X Labs (2017). *DNA Barcode Scanner*. Available at: https://conservationx.com/project/key/dnabarcoder (Accessed: 16 July 2018).

Conservation X Labs (2018). *Con X Tech Prize: Hacking Extinction*. Available at: https://conservationx.com/challenge/cxtech/prize (Accessed: 16 July 2018).

Cooke, B., Chudleigh, P., Simpson, S. and Saunders, G. (2013). 'The Economic Benefits of the Biological Control of Rabbits in Australia, 1950–2011'. *Australian Economic History Review*, 53(1):91–107. https://doi.org/10.1111/aehr.12000

Cooney, R., Kasterine, A., MacMillan, D., Milledge, S., Nossal, K., Roe, D. and Sas-Rolfes, M. (2015). *The trade in wildlife: a framework to improve biodiversity and livelihood outcomes*. International Trade Centre.

Courchamp, F., Chapuis, J.-L. and Pascal, M. (2003). 'Mammal invaders on islands: impact, control and control impact'. *Biological Reviews* 78(3).347–383. https://doi.org/10.1017/S1464793102006061

Cox, T., Strive, T., Mutze, G., West, P. and Saunders, G. (2013). *Benefits of Rabbit Biocontrol in Australia*. Canberra: PestSmart Toolkit publication, Invasive Animals Cooperative Research Centre.

Crawford, A.J., Lips, K.R. and Bermingham, E. (2010). 'Epidemic disease decimates amphibian abundance, species diversity, and evolutionary history in the highlands of central Panama'. *Proceedings of the National Academy of Sciences* 107(31):13777–13782. https://doi.org/10.1073/pnas.0914115107

Creel, L. (2003). *Ripple effects: population and coastal regions. Making the link*. Washington, D.C.: Population Reference Bureau.

Cross, M.L., Buddle, B.M. and Aldwell, F.E. (2007). 'The potential of oral vaccines for disease control in wildlife species'. *The Veterinary Journal* 174(3):472–480.

Crowl, T.A., Crist, T.O., Parmenter, R.R., Belovsky, G. and Lugo, A.E. (2008). 'The spread of invasive species and infectious disease as drivers of ecosystem change., *Frontiers in Ecology and the Environment* 6(5):238–246. https://doi.org/10.1890/070151

Crudge, B., Nguyen, T. and Cao, T.T. (2018). 'The challenges and conservation implications of bear bile farming in Viet Nam'. *Oryx*, pp. 1–8. https://doi.org/10.1017/S0030605317001752

Cuskelly, K. (2011). *Customs and Constitutions: State recognition of customary law around the world*. Bangkok, Thailand: IUCN. Available at: https://portals.iucn.org/library/node/10144

D'Amico, K.M., Horton, T.R., Maynard, C.A., Stehman, S.V., Oakes, A.D. and Powell, W.A. (2015). 'Comparisons of ectomycorrhizal colonization of transgenic American chestnut with those of the wild type, a conventionally bred hybrid, and related fagaceae species'. *Applied and Environmental Microbiology* 81(1):100–108. https://doi.org/10.1128/AEM.02169-14

Dabrock, P. (2009). 'Playing God? Synthetic biology as a theological and ethical challenge'. *Systems and Synthetic Biology* 3(1–4):47. https://doi.org/10.1007/s11693-009-9028-5

Dalia, T.N., Hayes, C.A., Stolyar, S., Marx, C.J., McKinlay, J.B. and Dalia, A.B. (2017). 'Multiplex genome editing by natural transformation (MuGENT) for synthetic biology in Vibrio natriegens'. *ACS Synthetic Biology*. ACS Publications, 6(9):1650–1655. https://doi.org/10.1021/acssynbio.7b00116

Damjanovic, K., Blackall, L.L., Webster, N.S. and van Oppen, M.J.H. (2017). 'The contribution of microbial biotechnology to mitigating coral reef degradation'. *Microbial biotechnology* 10(5):1236–1243. https://doi.org/10.1111/1751-7915.12769

Das, S. and Dash, H.R. (2017). *Handbook of Metal-microbe Interactions and Bioremediation*, 1st edition. CRC Press. https://doi.org/10.1201/9781315153353

De'ath, G., Fabricius, K.E., Sweatman, H. and Puotinen, M. (2012). 'The 27–year decline of coral cover on the Great Barrier Reef and its causes'. *Proceedings of the National Academy of Sciences* 109(44):17995–17999. https://doi.org/10.1073/pnas.1208909109

Dearden, P.K., Gemmell, N.J., Mercier, O.R., Lester, P.J., Scott, M.J., Newcomb, R.D., Buckley, T.R., Jacobs, J.M.E., Goldson, S.G. and Penman, D.R. (2017). 'The potential for the use of gene drives for pest control in New Zealand: a perspective'. *Journal of the Royal Society of New Zealand* 48(4):252–244.

Defense Advanced Research Projtects Agency (DARPA) (2016). *Research Announcement Young Faculty Award (YFA) DARPA-RA-16-63*. Available at: https://www.fbo.gov/utils/view?id=c1540b48aa08624b27f4dc5e7cdf94fe (Accessed: 16 July 2018).

Defense Advanced Research Projtects Agency (DARPA) (2018a). *Biological Robustness in Complex Settings (BRICS)*. Available at: https://www.darpa.mil/program/biological-robustness-in-complex-settings (Accessed: 1 August 2018).

Defense Advanced Research Projtects Agency (DARPA) (2018b). *Insect Allies*. Available at: https://www.darpa.mil/program/insect-allies (Accessed: 1 August 2018).

Defense Advanced Research Projtects Agency (DARPA) (2018c). *Living Foundries*. Available at: https://www.darpa.mil/program/living-foundries (Accessed: 16 July 2018).

Defense Advanced Research Projtects Agency (DARPA) (2018d). *Safe Genes*. Available at: https://www.darpa.mil/program/safe-genes (Accessed: 16 July 2018).

Delborne, J., Schneider, J., Bal, R., Cozzens, S. and Worthington, R. (2013). 'Policy pathways, policy networks, and citizen deliberation: Disseminating the results of World Wide Views on Global Warming in the USA'. *Science and Public Policy* 40(3):378–392. https://doi.org/10.1093/scipol/scs124

Delborne, J.A. (2008). 'Transgenes and transgressions: Scientific dissent as heterogeneous practice'. *Social Studies of Science* 38(4):509–541. https://doi.org/10.1177/0306312708089716

Delborne, J.A., Binder, A.R., Rivers, L., Barnes, J.C., Barnhill-Dilling, K., George, D., Kokotovich, A. and Sudweeks, J. (2018). *Biotechnology, the American Chestnut Tree, and Public Engagement (Workshop Report).* Available at: http://go.ncsu.edu/ges-chestnut-report

Department of Business Economic Development and Tourism (2004). *Planning for sustainable tourism*. Honolulu. Available at: http://files.hawaii.gov/dbedt/visitor/sustainable-tourism-project/drafts/General-Pop-Socio-Cultural-Report.pdf.

Deredec, A., Burt, A. and Godfray, C. (2008). 'The population genetics of using homing endonuclease genes (HEGs) in vector and pest management'. *Genetics* 179(4):2013–2026. https://doi.org/10.1534/genetics.108.089037

DeSalle, R. and Amato, G. (2004). 'The expansion of conservation genetics'. *Nature Reviews Genetics* 5(9):702. https://doi.org/10.1038/nrg1425

DeSalle, R. and Amato, G. (2017). 'Conservation Genetics, Precision Conservation, and De-extinction'. *The Hastings Center report* 47(Suppl 2):s18–s23. https://doi.org/10.1002/hast.747

Dhir, B. (2017). 'Bioremediation Technologies for the Removal of Pollutants'. In: R. Kumar, A. Sharma and S. Ahluwalia (ed.) *Advances in Environmental Biotechnology*, pp. 69–91. Springer, Singapore. https://doi.org/10.1007/978-981-10-4041-2_5

Dhole, S., Vella, M. R., Lloyd, A. L. and Gould, F. (2018). 'Invasion and migration of spatially self-limiting gene drives: A comparative analysis'. *Evolutionary Applications* 11(5):794–808. https://doi.org/10.1111/eva.12583

DiCastri, F. (1989). 'History of biological invasions with special emphasis on the Old World'. In: J.A. Drake, J. Mooney, F. DiCastri, R.H. Groves, F.J. Kruger, M. Rejmanek and M. Williamson (eds.) *Biological invasions: a global perspective*. Chichester, UK: Wiley-Blackwell.

Ding, J.L. and Ho, B. (2001). 'A new era in pyrogen testing.'. *Trends in Biotechnology* 19(8):277–81. https://doi.org/10.1016/S0167-7799(01)01694-8

Ding, J.L., Navas, M.A. and Ho, B. (1995). 'Molecular cloning and sequence analysis of factor C cDNA from the Singapore horseshoe crab, Carcinoscorpius rotundicauda'. *Molecular Marine Biology and Biotechnology* 4(1):90–103.

Dixon, G.B., Davies, S.W., Aglyamova, G.V., Meyer, E., Bay, L.K. and Matz, M.V. (2015). 'Genomic determinants of coral heat tolerance across latitudes'. *Science* 348(6242):1460–1462. https://doi.org/10.1126/science.1261224

DIYbio (2011). *Codes*. Available at: https://diybio.org/codes/ (Accessed: 25 July 2018).

Doherty, T.S., Glen, A.S., Nimmo, D.G., Ritchie, E.G. and Dickman, C.R. (2016). 'Invasive predators and global biodiversity loss'. *Proceedings of the National Academy of Sciences* 113(40):11261–11265. https://doi.org/10.1073/pnas.1602480113

Domergue, F., Abbadi, A. and Heinz, E. (2005). 'Relief for fish stocks: oceanic fatty acids in transgenic oilseeds'. *Trends in PlantSscience* 10(3):112–116. https://doi.org/10.1016/j.tplants.2005.01.003

Duckworth, J.W., Batters, G., Belant, J.L., Bennett, E.L., Brunner, J., Burton, J., Challender, D.W.S., Cowling, V., Duplaix, N., Harris, J.D., Hedges, S., Long, B., Mahood, P.J., McGowan, K., McShea, W.J., Oliver, W.L.R., Perkin, S., Rawson, B.M., Shepherd, C.R., Stuart, S.N., Talukdar, B.K., van Dijk, P.P., Vie, J-C., Walston, J.L., Whitten T. and Wirth, R. (2012). 'Why South-east Asia should be the world's priority for averting imminent species extinctions, and a call to join a developing cross-institutional programme to tackle this urgent issue'. *S.A.P.I.EN.S. Surveys and Perspectives Integrating Environment and Society* (5.2). Institut Veolia Environnement.

Duensing, N., Sprink, T., Parrott, W.A., Fedorova, M., Lema, M.A., Wolt, J.D. and Bartsch, D. (2018). 'Novel Features and Considerations for ERA and Regulation of Crops Produced by Genome Editing'. *Frontiers in Bioengineering and Biotechnology* 6(June):1–16. https://doi.org/10.3389/fbioe.2018.00079

Duke, S.O. (2003). 'Weeding with transgenes'. *Trends in Biotechnology* 21(5):192–195. https://doi.org/10.1016/S0167-7799(03)00056-8

Dulvy, N.K., Fowler, S.L., Musick, J.A., Cavanagh, R.D., Kyne, P.M., Harrison, L.R., Carlson, J.K., Davidson, L.N.K., Fordham, S.V., Francis, M.P., Pollock, C.M., Simpfendorfer, C.A., Burgess, G.H., Carpenter, K.E., Compagno, L.J.V., Ebert, D.A., Gibson, C., Heupel, M.R., Livingstone, S.R., Sanciangco, J.C., Stevens, J.D., Valenti, S. and White, W.T. (2014). 'Extinction risk and conservation of the world's sharks and rays.' https://doi.org/10.7554/eLife.00590 .

Dumroese, R.K., Williams, M.I., Stanturf, J.A. and St. Clair, J.B. (2015). 'Considerations for restoring temperate forests of tomorrow: forest restoration, assisted migration, and bioengineering'. *New Forests* 46(5–6):947–964. https://doi.org/10.1007/s11056-015-9504-6

Dvořák, P., Nikel, P.I., Damborský, J. and de Lorenzoa, V. (2017). 'Bioremediation 3.0: Engineering pollutant-removing bacteria in the times of systemic biology'. *Biotechnology Advances* 35(7):845–866. https://doi.org/10.1016/j.biotechadv.2017.08.001

Early, R., Bradley, B.A., Dukes, J.S., Lawler, J.J., Olden, J.D., Blumenthal, D.M., Gonzalez, P., Grosholz, E.D., Ibañez, I., Miller, L.P., Sorte, C.J.B. and Tatem, A.J. (2016). 'Global threats from invasive alien species in the twenty-first century and national response capacities'. *Nature Communications* 7:12485. https://doi.org/10.1038/ncomms12485

Eason, C.T., Shapiro, L., Ogilvie, S., King, C. and Clout, M. (2017). 'Trends in the development of mammalian pest control technology in New Zealand'. *New Zealand Journal of Zoology* 44(4):267–304. https://doi.org/10.1080/03014223.2017.1337645

Eaton, J.A., Shepherd, C.R., Rheindt, F.E., Harris, J.B.C., Balen, B., Wilcove, D.S. and Collar, N.J. (2015). 'Trade-driven extinctions and near-extinctions of avian taxa in Sundaic Indonesia'. *Forktail* (31):1–12.

Eckhoff, P.A., Wenger, E.A., Godfray, H.C.J. and Burt, A. (2017). 'Impact of mosquito gene drive on malaria elimination in a computational model with explicit spatial and temporal dynamics'. *Proceedings of the National Academy of Sciences* 114(2):E255–E264. https://doi.org/10.1073/pnas.1611064114

Eden, G. (2014). 'Special Eurobarometer 401: survey summary on responsible research and innovation, science and technology'. *Journal of Responsible Innovation* 1(1):129–132. https://doi.org/10.1080/23299460.2014.882553

Ehrenfeld, J.G. (2010). 'Ecosystem consequences of biological invasions'. *Annual Review of Ecology, Evolution, and Systematics* 41:59–80. https://doi.org/10.1146/annurev-ecolsys-102209-144650

Ehrlich, P.R. and Ehrlich, A.H. (2014). *The case against de-extinction: it's a fascinating but dumb idea*, *Yale Environment 360*. Available at: https://e360.yale.edu/features/the_case_against_de-extinction_its_a_fascinating_but_dumb_idea (Accessed: 18 July 2018).

Elliott, K.C. (2013): 'Selective ignorance and agricultural research'. *Science, Technology, & Human Values* 38(3):328–350. https://doi.org/10.1177/0162243912442399

Emerson, C., James, S., Littler, K. and Randazzo, F.F. (2017). 'Principles for gene drive research'. *Science* 358(6367):1135–1136. https://doi.org/10.1126/science.aap9026

ERASynBio (2014). *Next Steps for European Synthetic Biology: a strategic vision from ERASynBio*.

Erosion, Technology and Concentration (ETC) Group (2013). *Case Study: Vanilla and Synthetic Biology*. Available at: http://www.etcgroup.org/content/case-study-vanilla (Accessed: 16 July 2018).

Erosion, Technology and Concentration (ETC) Group (2018). *Synthetic biology*. Available at: http://www.etcgroup.org/issues/synthetic-biology

Escaler, M., Teng, P.P.S. and Powell, A.D. (2012). 'Challenges of Harmonization of Agricultural Biotechnology Regulatory Systems across APEC Economies'. *Biosafety* 01(02). https://doi.org/10.4172/2167-0331.1000101

Estes, J.A., Terborgh, J., Brashares, J.S., Power, M.E., Berger, J., Bond, W.J., Carpenter, S.R., Essington, T.E., Holt, R.D., Jackson, J.B.C., Marquis, R.J., Oksanen, L., Oksanen, T., Paine, R.T., Pikitch, E.K., Ripple, W.J., Sandin, S.A., Scheffer, M., Schoener, T.W., Shurin, J.B., Sinclair, A.R.E., Soulé, M.E., Virtanen, R. and Wardle, D.A. (2011). 'Trophic downgrading of planet Earth'. *Science* 333(6040):301–306. https://doi.org/10.1126/science.1205106

Estes, J.A., Burdin, A. and Doak, D.F. (2016). 'Sea otters, kelp forests, and the extinction of Steller's sea cow'. *Proceedings of the National Academy of Sciences* 113(4):880–885. https://doi.org/10.1073/pnas.1502552112

Esvelt, K., Smidler, A.L., Catteruccia, F. and Church, G.M. (2014). 'Concerning RNA-guided gene drives for the alteration of wild populations. eLIFE (art. e03401)'. https://doi.org/10.7554/eLife.03401

European Academies' Science Advisory Council (EASAC) (2017). *Genome editing: scientific opportunities, public interests and policy options in the European Union.* Halle/Saale. Available at: https://easac.eu/fileadmin/PDF_s/reports_statements/Genome_Editing/EASAC_Report_31_on_Genome_Editing.pdf.

European Environmental Agency (EEA) (2013). *Late lessons from early warnings: science, precaution, innovation*. Available at: https://www.eea.europa.eu/publications/late-lessons-2.

European Food Safety Authority (EFSA) (2010). *EFSA Panel on Genetically Modified Organisms (GMO), Guidance on the environmental risk assessment of genetically modified plants*.

European Food Safety Authority (EFSA) (2018). 'Guidelines on EFSA Consultations'. *EFSA Supporting Publications* 15(3):1390E.

European Food Safety Authority (EFSA) Scientific Committee (2017). 'Guidance on the use of the weight of evidence approach in scientific assessments'. *EFSA Journal* 15(8):e04971.

European Network of Scientists for social and environmental responsibility (ENSSER) (2017). *Statement on New Genetic Modification Techniques*. Available at: https://ensser.org/publications/ngmt-statement/.

Evolva (2018). *Vanillin*. Available at: https://www.evolva.com/vanillin/ (Accessed: 1 June 2018).

Ewen, J.G. (2012). *Reintroduction biology: integrating science and management*. John Wiley & Sons. https://doi.org/10.1002/9781444355833

Federal Register (1978). *Federal Register: 42 Fed. Reg. 57683 (Nov. 4, 1977).*

Feil, R. and Fraga, M.F. (2012). 'Epigenetics and the environment: emerging patterns and implications'. *Nature reviews genetics* 13(2):97.

Ferraro, P.J. and Pattanayak, S.K. (2006). 'Money for Nothing? A Call for Empirical Evaluation of Biodiversity Conservation Investments'. *PLoS Biology* 4(4):e105. https://doi.org/10.1371/journal.pbio.0040105

Feschotte, C. and Pritham, E.J. (2007). 'DNA Transposons and the Evolution of Eukaryotic Genomes'. *Annual Review of Genetics* 41(1):331–368. https://doi.org/10.1146/annurev.genet.40.110405.090448

Foundation for the National Institutes of Health (FNIH) (2018a). *Gene Drive Research Consortium*. Available at: https://fnih.org/what-we-do/current-research-programs/gene-drive-research-consortium (Accessed: 25 July 2018).

Foundation for the National Institutes of Health (FNIH) (2018b). 'Gene Drive Research Sponsors and Supporters Forum'. Available at: https://fnih.org/what-we-do/current-lectures-awards-and-events/gene-drive-research-forum.

Finless Foods (2018). *Finless Foods*.

Fisher, E.C., Jones, J.S. and von Schomberg, R. (2006). *Implementing the precautionary principle: perspectives and prospects*. Edward Elgar Publishing. https://doi.org/10.4337/9781847201676.00009

Fisher, M.C., Henk, D.A., Briggs, C.J., Brownstein, J.S., Madoff, L.C., McCraw, S.L. and Gurr, S.J. (2012). 'Emerging fungal threats to animal, plant and ecosystem health'. *Nature* 484:186–194. https://doi.org/10.1038/nature10947

Fisher, M.C. and Garner, T.W. (2007). 'The relationship between the emergence of Batrachochytrium dendrobatidis, the international trade in amphibians and introduced amphibian species'. *Fungal Biology Reviews* 21:2–9. https://doi.org/10.1016/j.fbr.2007.02.002

Fisher, M.C., Garner, T.W.J. and Walker, S.F. (2009). 'Global emergence of Batrachochytrium dendrobatidis and amphibian chytridiomycosis in space, time, and host'. *Annual Review of Microbiology* 63:291–310. https://doi.org/10.1146/annurev.

micro.091208.073435

Fister, A.S., Landherr, L., Maximova, S.N. and Guiltinan, M.J. (2018). 'Transient expression of CRISPR/Cas9 machinery targeting TcNPR3 enhances defense response in Theobroma cacao'. *Frontiers in Plant Science* 9:268. https://doi.org/10.3389/fpls.2018.00268

Flajnik, M.F. (2018). 'A cold-blooded view of adaptive immunity'. *Nature Reviews Immunology* 18:438–453. https://doi.org/10.1038/s41577-018-0003-9

Fleischer, R.C., James, H.F. and Olson, S.L. (2008). 'Convergent Evolution of Hawaiian and Australo-Pacific Honeyeaters from Distant Songbird Ancestors'. *Current Biology* 18(24):1927–1931. https://doi.org/10.1016/J.CUB.2008.10.051

Foley, J.A., DeFries, R., Asner, G.P., Barford, C., Bonan, G., Carpenter, S.R., Chapin, F.S., Coe, M.T., Daily, G.C., Gibbs, H.K., Helkowski, J.H., Holloway, T., Howard, E.A., Kucharik, C.J., Monfreda, C., Patz, J.A., Prentice, C., Ramankutty, N. and Snyder, P.K. (2005). 'Global consequences of land use'. *Science* 309(5734):570–574. https://doi.org/10.1126/science.1111772

Fontaine, M.C., Pease, J.B., Steele, A., Waterhouse, R.M., Neafsey, D.E., Sharakhov, I.V., Jiang, X., Hall, A.B., Catteruccia, F., Kakani, E., Mitchell, S.N., Wu, Y.-C., Smith, H.A., Love, R.R., Lawniczak, M.K., Slotman, M.A., Emrich, S.J., Hahn, M.W. and Besansky, N.J. (2015). 'Extensive introgression in a malaria vector species complex revealed by phylogenomics'. *Science* 347(6217). https://doi.org/10.1126/science.1258524

Food and Agriculture Organization of the United Nations (FAO) (2017). *The State of food and agriculture: Leveraging food systems for inclusive rural transformation*. https://doi.org/10.18356/1a078735-en

Food and Agriculture Organization of the United Nations (FAO) (2016). *Food and Agriculture: Key to achieving the 2030 Agenda for Sustainable Development*.

Food and Drug Administration (FDA) (2017a). *Clarification of FDA and EPA Jurisdiction over Mosquito-Related Products*. United States.

Food and Drug Administration (FDA) (2017b). *Regulation of intentionally altered genomic DNA in animals*. United States.

Fortini, L.B., Vorsino, A.E., Amidon, F.A., Paxton, E.H. and Jacobi, J.D. (2015). 'Large-scale range collapse of Hawaiian forest birds under climate change and the need 21st century conservation options'. *PloS one* 10(10):e0140389. https://doi.org/10.1371/journal.pone.0144311

Foster, A. and Pummill, A. (2011). 'Saving the Honeybees: A Synthetic Biology Approach'. Missouri University of Science and Technology.

Francis (2015). Laudato Si: *On Care for our Common Home*. Encyclial Letter doi: 10.1017/S000358150009199X

Frankham, R. (2015). 'Genetic rescue of small inbred populations: meta-analysis reveals large and consistent benefits of gene flow'. *Molecular Ecology* 24(11):2610–2618. https://doi.org/10.1111/mec.13139

Friends of the Earth (FOE) (2012). 'The Principles for the Oversight of Synthetic Biology'. *Elsevier*, pp. 1–20. Available at: www.foe.org/news/blog/2012-03-global-coalition-calls-oversight-synthetic-biology.

Fritsche, S., Poovaiah, C., MacRae, E. and Thorlby, G. (2018). 'A New Zealand Perspective on the Application and Regulation of Gene Editing'. *Frontiers in Plant Science* 9:1323. https://doi.org/10.3389/fpls.2018.01323

Fuentes-Pardo, A.P. and Ruzzante, D.E. (2017). 'Whole-genome sequencing approaches for conservation biology: Advantages, limitations and practical recommendations'. *Molecular Ecology* 26(20):5369–5406. https://doi.org/10.1111/mec.14264

Fuller, L., Marzano, M., Peace, A., Quine, C.P. and Dandy, N. (2016). 'Public acceptance of tree health management: Results of a national survey in the UK'. *Environmental Science & Policy* 59:18–25. https://doi.org/10.1016/j.envsci.2016.02.007

Gales, N. (2011). 'Humpback whales: status in the southern hemisphere'. *Journal of Cetacean Research and Management* (Special Issue 3). Available at: https://archive.iwc.int/?c=28 International Whaling Commission.

Galloway, T.S., Cole, M. and Lewis, C. (2017). 'Interactions of microplastic debris throughout the marine ecosystem'. *Nature ecology & evolution* 1(5):116. https://doi.org/10.1038/s41559-017-0116

Gallup, J. and Sachs, J. (2001). The economic burden of malaria. Supplement to Volume 64(1) of the *American Journal of Tropical Medicine and Hygiene* 64(1):85–96. https://doi.org/10.4269/ajtmh.2001.64.85

Gannon, F. (2001). 'The essential role of peer review'. *EMBO reports* 2(9):743.

Gantz, V.M., Jasinskiene, N., Tatarenkova, O., Fazekas, A., Macias, V.M., Bier, E. and James, A.A. (2015). 'Highly efficient Cas9-mediated gene drive for population modification of the malaria vector mosquito *Anopheles stephensi*'. *Proceedings of the National*

Academy of Sciences 112(49):E6736–E6743. https://doi.org/10.1073/pnas.1521077112

Garnett, S.T., Garnett, S.T., Burgess, N.D., Fa, J.E., Fernández-Llamazares, A., Molnár, Z., Robinson, C.J., Watson, J.E.M., Zander, K.K., Austin, B., Brondizio, E.S., Collier, N.F., Duncan, T., Ellis, E., Geyle, H., Jackson, M.V., Jonas, H., Malmer, P., McGowan, B., Sivongxay, A. and Leiper, I. (2018). 'A spatial overview of the global importance of Indigenous lands for conservation'. *Nature Sustainability* 1(7):369–374. https://doi.org/10.1038/s41893-018-0100-6

Garrett, L. (2013). 'Biology's Brave New World: The Promise and Perils of the Synbio Revolution'. *Foreign Affairs*, 92(6):28–46.

Gasco, L., Gai, F., Maricchiolo, G., Genovese, L., Ragonese, S., Bottari, T. and Caruso, G. (2018). 'Sustainable Alternatives for Dietary Fish Oil in Aquafeeds: Actual Situation and Future Perspectives'. In: *Feeds for the Aquaculture Sector*, pp. 49–61. Springer. https://doi.org/10.1007/978-3-319-77941-6_3

Gauvry, G. (2015). 'Current Horseshoe Crab Harvesting Practices Cannot Support Global Demand for TAL/LAL: The Pharmaceutical and Medical Device Industries' Role in the Sustainability of Horsehose Crabs' (chapter 7). In: *Changing Global Perspectives on Horseshoe Crab Biology, Conservation and Management*, pp. 1–599. https://doi.org/10.1007/978-3-319-19542-1_27

Gemmell, N.J. and Tompkins, D.M. (2017). 'Gene drives and rodent control: response to Piaggio et al.'. *Trends in Ecology & Evolution* 32(5):314–315. https://doi.org/10.1016/j.tree.2017.03.005

Genz, A., Brinck, K.W., Berkowitz, P.S. and Jacobi, J.D. (2018). *2017-2018 Palila abundance estimates and trend. Hawai'i Cooperative Studies Unit Technical Report*. Hilo.

German Central Commission of Biological Safety (GCCBS) (2016). *Position statement of the ZKBS on the classification of genetic engineering operations for the production and use of higher organisms using recombinant gene drive systems*. Available at: https://bch.cbd.int/database/record.shtml?documentid=110745

Ghana EPA, Environment Protection Agency Ghana (2003). *Annual report: revised draft*. Accra, Ghana.

Gieryn, T.F. (1999). *Cultural boundaries of science: Credibility on the line*. University of Chicago Press.

Gilpin, M.E. and Soulé, M.E. (no date). 'Minimum Viable Populations: Processes of Species Extinction'. In: M.E. Soulé (ed.) *Conservation Biology: The Science of Scarcity and Diversity*, pp. 19–34. Sunderland, Mass: Sinauer.

Glen, A.S., Atkinson, R., Campbell, K.J., Hagen, E., Holmes, N.D., Keitt, B.S., Parkes, J.P., Saunders, A., Sawyer, J. and Torres, H. (2013). 'Eradicating multiple invasive species on inhabited islands: the next big step in island restoration?'. *Biological Invasions* 15(12):2589–2603. https://doi.org/10.1007/s10530-013-0495-y

Globe Newswire (2018). *Synthetic Biology Global Markets to Reach $13.9 Billion by 2022*. Available at: https://globenewswire.com/news-release/2018/07/11/1535887/0/en/synthetic-biology-global-markets-to-reach-13-9-billion-by-2022.html (Accessed: 16 July 2018).

GMO-Free Europe (2016). *'Germany' (GMO Free Regions by Country, 2016)*. Available at: https://www.gmo-free-regions.org/gmo-free-regions/germany.html (Accessed: 25 July 2018). https://doi.org/10.1080/13669877.2015.1034161

Goldspiel, H., Newhouse, A.E., Powell, W.A. and Gibbs, J.P. (2018). 'Effects of Transgenic American Chestnut Leaf Litter on Growth and Survival of Wood Frog Larvae'. *Restoration Ecology* 27(2):371–378. https://doi.org/10.1111/rec.12879

Goldstein, B., Moses, R., Sammons, N. and Birkved, M. (2017). 'Potential to curb the environmental burdens of American beef consumption using a novel plant-based beef substitute'. *PloS one*, 12(12). https://doi.org/10.1371/journal.pone.0189029

Gomez, M.A., Lin, Z.D., Moll, T., Chauhan, R.D., Hayden, L., Renninger, K., Beyene, G., Taylor, N.J., Carrington, J.C., Staskawicz, B.J. and Bart, R.S. (2018). 'Simultaneous CRISPR/Cas9-mediated editing of cassava eIF4E isoforms nCBP-1 and nCBP-2 reduces cassava brown streak disease symptom severity and incidence'. Plant Biotechnology Journal 17(2):421-434. https://doi.org/10.1111/pbi.12987

Gong, W., Sinden, J., Braysher, M. and Jones, R. (2009). *The economic impacts of vertebrate pests in Australia*.

Good, A. (2018). 'Toward nitrogen-fixing plants'. *Science* 359(6378):869–870. https://doi.org/10.1126/science.aas8737

Gorresen, P.M., Camp, R.J., Reynolds, M.H., Woodworth, B.L. and Pratt, T.K. (2009). 'Status and trends of native Hawaiian songbirds'. In: T.K. Pratt, C.T. Atkinson, P.C. Banko, J.D. Jacobi and B.L. Woodworth (eds.) *Conservation biology of Hawaiian forest birds: implications for island avifauna*, pp. 108–136. New Haven, Connecticut, USA: Yale University Press. https://doi.org/10.1525/auk.2010.127.4.956

Gratwicke, B., Bennett, E.L., Broad, S., Christie, S., Dutton, A., Gabriel, G., Kirkpatrick, C. and Nowell, K. (2008). 'The world can't have

wild tigers and eat them, too'. *Conservation Biology* 22(1):222–223. https://doi.org/10.1111/j.1523-1739.2007.00802.x

Greenspan, S.E., Lambertini, C., Carvalho, T., James, T.Y., Toledo, L.F., Haddad, C.F.B. and Becker, C.G. (2018). 'Hybrids of amphibian chytrid show high virulence in native hosts'. *Scientific Reports* 8(1):9600. https://doi.org/10.1038/s41598-018-27828-w

Gressel, J. (2002). *Molecular biology of weed control*. CRC Press.

Grewal, D.S. (2017). 'Before Peer Production: Infrastructure Gaps and the Architecture of Openness in Synthetic Biology'. *Stanford Technology Law Review* 20(1):143.

Gross, R.E. (2014). *Holy Cow: Would lab-grown meat ever be kosher? Slate2*. Available at: http://www.slate.com/articles/technology/future_tense/2014/09/lab_grown_meat_is_it_kosher.html.

Grunwald, H.A., Gantz, V.M., Poplawski, G., Xu, X.S., Bier, E. and Cooper, K.L. (2018). 'Super-Mendelian inheritance mediated by CRISPR/Cas9 in the female mouse germline'. *bioRxiv*. https://doi.org/10.1101/362558

Grushkin, D., Kuiken, T. and Millet, P. (2013). *Seven Myths & Realities about Do-It-Yourself Biology*. Washington, D.C.

Guan, Z., Schmidt, M., Pei, L., Wei, W. and Ma, K. (2013). 'Biosafety Considerations of Synthetic Biology in the International Genetically Engineered Machine (iGEM) Competition'. *BioScience* 63(1):25–34. https://doi.org/10.1525/bio.2013.63.1.7

Gumulya, Y., Boxall, N., Khaleque, H.N., Santala, V., Carlson, R.P. and Kaksonen, A.H. (2018). 'In a quest for engineering acidophiles for biomining applications: challenges and opportunities'. *Genes*. Multidisciplinary Digital Publishing Institute, 9(2):116. https://doi.org/10.3390/genes9020116

Gustafson, E.J., de Bruijn, A., Lichti, N., Jacobs, D.F., Sturtevant, B.R., Foster, J., Miranda, B.R. and Dalgleish, H.J. (2017). 'The implications of American chestnut reintroduction on landscape dynamics and carbon storage'. *Ecosphere* 8(4):e01773.

Hagiwara, D., Takahashi, H., Watanabe, A., Takahashi-Nakaguchi, A., Kawamoto, S., Kamei, K. and Gonoi, T. (2014). 'Whole-Genome Comparison of *Aspergillus fumigatus* Strains Serially Isolated from Patients with Aspergillosis'. *Journal of Clinical Microbiology* 52(12):4202–4209. https://doi.org/10.1128/jcm.01105-14

Hairston, N.A. and Hairston, N.G. (1987). *Community ecology and salamander guilds*. Cambridge University Press.

Hajjar, R. and Kozak, R.A. (2015). 'Exploring public perceptions of forest adaptation strategies in Western Canada: Implications for policy-makers'. *Forest Policy and Economics* 61:59–69.

Hale, K.A. and Briskie, J.V (2007). 'Decreased immunocompetence in a severely bottlenecked population of an endemic New Zealand bird'. *Animal Conservation* 10(1):2–10. https://doi.org/10.1111/j.1469-1795.2006.00059.x

Hall, M.R., Kocot, K.M., Baughman, K.W., Fernandez-Valverde, L., Gauthier, M.E.A., Hatleberg, W.L., Krishnan, A., McDougall, C., Motti, C.A., Shoguchi, E., Wang, T., Xiang, X., Zhao, M., Bose, U., Shinzato, C., Hisata, K., Fujie, M., Kanda, M., Cummins, S.F., Satoh, N., Degnan, S.M. and Degnan, B.M. (2017). 'The crown-of-thorns starfish genome as a guide for biocontrol of this coral reef pest'. *Nature* 544:231–234. https://doi.org/10.1038/nature22033

Hamdan, M.N., Post, M., Ramli, M.A. and Mustafa, A.R. (2018). 'Cultured Meat in Islamic Perspective'. *Journal of Religion and Health*, 57(6):2193–2206. https://doi.org/10.1007/s10943-017-0403-3

Hammitt, J.K., Wiener, J.B., Swedlow, B., Kall, D. and Zhou, Z. (2005). 'Precautionary regulation in Europe and the United States: a quantitative comparison'. *Risk Analysis: An International Journal* 25(5):1215–1228. https://doi.org/10.1111/j.1539-6924.2005.00662.x

Hammond, A.M., Kyrou, K., Bruittini, M., North, A., Galizi, R., Karlsson, X., Kranjc, N., Carpi, F.M., D'Aurizio, R., Crisanti, A. and Nolan, T. (2017). 'The creation and selection of mutations resistant to a gene drive over multiple generations in the malaria mosquito'. *PLoS Genetics*, 13(10):1–16. https://doi.org/10.1371/journal.pgen.1007039

Hanson, J. (2014). 'Precautionary principle'. *Bioethics*. 4th edition. Macmillan Reference.

Harremoës, P., Gee, D., MacGarvin, M., Stirling, A., Keys, J., Wynne, B. and Vaz, S.G. (2002). *Late lessons from early warnings: the precautionary principle 1896-2000*. Available at: https://www.eea.europa.eu/publications/environmental_issue_report_2001_22. https://doi.org/10.4324/9781315071985

Harris, R.N., Brucker, R.M., Walke, J.B., Becker, M.H., Schwantes, C.R., Flaherty, D.C., Lam, B.A., Woodhams, D.C., Briggs, C.J., Vredenburg, V.T. and Minbiole, K.P.C. (2009). 'Skin microbes on frogs prevent morbidity and mortality caused by a lethal skin fungus'. *The ISME Journal* 3(7):818. https://doi.org/10.1038/ismej.2009.27

Harrison, S.T.L. (2016). 'Biotechnologies that Utilise Acidophiles'. In: *Acidophiles: Life in Extremely Acidic Environments*, pp. 265–284. Caister Academic Press. https://doi.org/10.21775/9781910190333.16

Hartley, S. and Kokotovich, A. (2017). 'Disentangling risk assessment: new roles for experts and publics'. *Science and the politics of openness*, p. 176. https://doi.org/10.7765/9781526106476.00019

Harvey-Samuel, T., Ant, T. and Alphey, L. (2017). 'Towards the genetic control of invasive species'. *Biological Invasions* 19(6):1683–1703. https://doi.org/10.1007/s10530-017-1384-6

Haseloff Lab, University of Cambridge (2018). *Synthetic Biology Reports*. Available at: http://www.haseloff-lab.org/synbotany/reports/index.html (Accessed: 25 June 2018).

Hayes, K. (2011). 'Uncertainty and uncertainty analysis methods'. CSIRO.

Hayes, K.R., Hosack, G.R., Dana, G.V., Foster, S.D., Ford, J.H., Thresher, R., Ickowicz, A., Peel, D., Tizard, M., De Barro, P., Strive, T. and Dambacher, J.M. (2018). 'Identifying and detecting potentially adverse ecological outcomes associated with the release of gene-drive modified organisms'. *Journal of Responsible Innovation* 5(sup1):S139–S158. https://doi.org/10.1080/23299460.2017.1415585

Heavey, P. (2017). 'Synthetic Biology: The Response of the Commission of the (Catholic) Bishops' Conferences of the European Community'. *Cambridge Quarterly of Healthcare Ethics* 26(2):257–266. https://doi.org/10.1017/s0963180116000852

Hebert, P.D.N., Cywinska, A. and Ball, S.L. (2003). 'Biological identifications through DNA barcodes'. *Proceedings of the Royal Society of London B: Biological Sciences* 270(1512):313–321. https://doi.org/10.1098/rspb.2002.2218

Hedrick, P.W. and Garcia-Dorado, A. (2016). 'Understanding inbreeding depression, purging, and genetic rescue'. *Trends in Ecology & Evolution* 31(12):940–952. https://doi.org/10.1016/j.tree.2016.09.005

Hickerson, C.-A.M., Anthony, C.D. and Walton, B.M. (2017). 'Eastern Red-backed Salamanders regulate top-down effects in a temperate forest-floor community'. *Herpetologica* 73(3):180–189. https://doi.org/10.1655/herpetologica-d-16-00081.1

Higg Materials Sustainability Index (2018). *Higg Materials Sustainability Index*. Available at: https://msi.higg.org/terms-of-use (Accessed: 16 July 2018).

Hilbeck, A., Meier, M., Römbke, J., Jänsch, S., Teichmann, H. and Tappeser, B. (2011). 'Environmental risk assessment of genetically modified plants - concepts and controversies'. *Environmental Sciences Europe* 23(1):13. https://doi.org/10.1186/2190-4715-23-13

Hildebrandt, T.B., Hermes, R>, Colleoni, S., Diecke, S., Holtze, S., Renfree, M.B., Stejskal, J., Hayashi, K., Drukker, M., Loi, P., Goritz, F., Lazzari, G. and Galli, C. (2018). 'Embryos and embryonic stem cells from the white rhinoceros'. *Nature Communications* 9(1):2589.

Hill, J., Copse, C., Leary, S., Stagg, A.J., Williamson, D. and Titball, R.W. (2003). 'Synergistic protection of mice against plague with monoclonal antibodies specific for the F1 and V antigens of Yersinia pestis'. *Infection and Immunity* 71(4):2234–2238. https://doi.org/10.1128/iai.71.4.2234-2238.2003

Hites, R.A., Foran, J.A., Carpenter, D.O., Hamilton, M.C., Knuth, B.A. and Schwager, S.J. (2004). 'Global assessment of organic contaminants in farmed salmon'. *Science*. 303(5655):226–229. https://doi.org/10.1126/science.1091447

Hixson, S.M., Shukla, K., Campbell, L.G., Hallett, R.H., Smith, S.M., Packer, L. and Arts, M.T. (2016). 'Long-chain omega-3 polyunsaturated fatty acids have developmental effects on the crop pest, the cabbage white butterfly Pieris rapae'. *PloS one*. 11(3):e0152264. https://doi.org/10.1371/journal.pone.0152264

Hoffman, E., Hanson, J. and Thomas, J. (2012). *The Principles for the Oversight of Synthetic Biology*.

Hoffmann, A.A., Montgomery, B.L., Popovici, J., Iturbe-Ormaetxe, I., Johnson, P.H., Muzzi, F., Greenfield, M., Durkan, M., Leong, Y.S., Dong, Y., Cook, H., Axford, J., Callahan, A.G., Kenny, N., Omodei, C., McGraw, E.A., Ryan, P.A., Ritchie, S.A., Turelli, M. and O'Neill, S.L. (2011). 'Successful establishment of Wolbachia in Aedes populations to suppress dengue transmission'. *Nature* 476:454–457. https://doi.org/10.1038/nature10356

Hoffmann, M., Hilton-Taylor, C., Angulo, A., Böhm, M., Brooks, T.M., Butchart, S.H.M., Carpenter, K.E., Chanson, J., Collen, B., Cox, N.A., Darwall, W.R.T., Dulvy, N.K., Harrison, L.R., Katariya, V., Pollock, C.M., Quader, S., Richman, N.I., Rodrigues, A.S.L., Tognelli, M.F., Vié, J., Aguiar, J.M., Allen, D.J., Allen, G.R., Amori, G., Ananjeva, N.B., Andreone, F., Andrew, P., Ortiz, A.L.A., Baillie, J.E.M., Baldi, R., Bell, B.D., Biju, S.D., Bird, J.P., Black-Decima, P., Blanc, J.J., Bolaños, F., Bolivar-G, W., Burfield, I.J., Burton, J.A., Capper, D.R., Castro, F., Catullo, G., Cavanagh, R.D., Channing, A., Chao, N.L., Chenery, A.M., Chiozza,

F., Clausnitzer, V., Collar, N.J., Collett, L.C., Collette, B.B., Fernandez, C.F.C., Craig, M.T., Crosby, M.J., Cumberlidge, N., Cuttelod, A., Derocher, A.E., Diesmos, A.C., Donaldson, J.S., Duckworth, J.W., Dutson, G., Dutta, S.K., Emslie, R.H., Farjon, A., Fowler, S., Freyhof, J., Garshelis, D.L., Gerlach, J., Gower, D.J., Grant, T.D., Hammerson, G.A., Harris, R.B., Heaney, L.R., Hedges, S.B., Hero, J., Hughes, B., Hussain, S.A., Icochea M., J., Inger, R.F., Ishii, N., Iskandar, D.T., Jenkins, R.K.B., Kaneko, Y., Kottelat, M., Kovacs, K.M., Kuzmin, S, L., La Marca, E., Lamoreux, J.F., Lau, M.W.N., Lavilla, E.O., Leus, K., Lewison, R.L., Lichtenstein, G., Livingstone, S.R., Lukoschek, V., Mallon, D.P., McGowan, P.J.K., McIvor, A., Moehlman, P.D., Molur, S., Alonso, A.M., Musick, J.A., Nowell, K., Nussbaum, R.A., Olech, W., Orlov, N.L., Papenfuss, T.J., Parra-Olea, G, Perrin, W.F., Polidoro, B.A., Pourkazemi, M., Racey, P.A., Ragle, J.S., Ram, M., Rathbun, G., Reynolds, R.P., Rhodin, A.G.J., Richards, S.J., Rodríguez, L.O., Ron, S.R., Rondinini, C., Rylands, A.B., de Mitcheson, Y.S., Sanciangco, J.C., Sanders, K.L., Santos-Barrera, G., Schipper, J., Self-Sullivan, C., Shi, Y, Shoemaker, A., Short, F.T., Sillero-Zubiri, C., Silvano, D.L., Smith, K.G., Smith, A.T., Snoeks, J., Stattersfield, A.J., Symes, A.J., Taber, A.B., Talukdar, B.K., Temple, H.J., Timmins, R., Tobias, J.A., Tsytsulina, K., Tweddle, D., Ubeda, C., Valenti, S.V., van Dijk, P.P., Veiga, L.M., Veloso, A., Wege, D.C., Wilkinson, M., Williamson, E.A., Xie, F., Young, B.E., Akçakaya, H.R., Bennun, L., Blackburn, T.M., Boitani, L., Dublin, H.T., da Fonseca, G.A.B., Gascon, C., Lacher Jr., T.E., Mace, G.M., Mainka, S.A., McNeely, J.A., Mittermeier, R.A., Reid, G.M., Rodriguez, J.P., Rosenberg, A.A., Samways, M.J., Smart, J., Stein, B.A. and Stuart, S. (2010). 'The impact of conservation on the status of the world's vertebrates'. *Science* 330(6010):1503–1509. https://doi.org/10.1126/science.1194442

Holman, C.M. (2014). 'Developments in synthetic biology are altering the IP imperatives of biotechnology'. *Vanderbilt J. Ent. & Tech. L.* 17:385.

Holtzman, L. and Gersbach, C.A. (2018). 'Editing the Epigenome: Reshaping the Genomic Landscape'. *Annual Review of Genomics and Human Genetics* 19:43–71. https://doi.org/10.1146/annurev-genom-083117-021632

Hooper, D.U., Adair, E.C., Cardinale, B.J., Byrnes, J.E., Hungate, B.A., Matulich, K.L., Gonzalez, A., Duffy, J.E., Gamfeldt, L. and O'Connor, M.I. (2012). 'A global synthesis reveals biodiversity loss as a major driver of ecosystem change'. *Nature* 486(7401):105.

Hopper, K.R., Britch, S.C. and Wajnberg, E. (2006). 'Risks of interbreeding between species used in biological control and native species, and methods for evaluating their occurrence and impact'. In: F. Bigler, D. Babendreier and U. Kuhlmann (eds.) *Environmental impact of invertebrates for biological control of arthropods: methods and risk assessment*, pp. 78–97. Wallingford: CABI. https://doi.org/10.1079/9780851990583.0078

Hou, L., Cui, Y., Li, X., Chen, W., Zhang, Z., Pang, X. and Li, Y. (2018). 'Genetic Evaluation of Natural Populations of the Endangered Conifer Thuja koraiensis Using Microsatellite Markers by Restriction-Associated DNA Sequencing'. *Genes* 9(4):218. https://doi.org/10.3390/genes9040218

Howald, G., Donlan, C.J., Galvan, J.P., Russell, J.C., Parkes, J., Samaniego, A., Wang, Y., Veitch, D., Genovesi, P., Pascal, M., Saunders, A. and Tershy, B. (2007). 'Invasive rodent eradication on islands'. *Conservation Biology* 21(5):1258–1268.

Howard, J.G., Lynch, C., Santymire, R.M., Marinari, P.E. and Wildt, D.E. (2016). 'Recovery of gene diversity using long-term cryopreserved spermatozoa and artificial insemination in the endangered black-footed ferret'. *Animal Conservation* 19(2):102–111. https://doi.org/10.1111/acv.12229

Hughes, T.P., Anderson, K.D., Connolly, S.R., Heron, S.F., Kerry, J.T., Lough, J.M., Baird, A.H., Baum, J.K., Berumen, M.L., Bridge, T.C., Claar, D.C., Eakin, C.M., Gilmour, J.P., Graham, N.A.J., Harrison, H., Hobbs, J.A., Hoogenboom, M., Lowe, R.J., McCulloch, M.T., Pandolfi, J.M., Pratchett, M., Schoepf, V., Torda, G. and Wilson, S.K. (2018). 'Spatial and temporal patterns of mass bleaching of corals in the Anthropocene'. *Science* 359(6371):80–83.

Hulme, P.E. (2014). 'Invasive species challenge the global response to emerging diseases'. *Trends in Parasitology* 30(6):267–270. https://doi.org/10.1016/j.pt.2014.03.005

Hunter, P. (2016). 'The potential of molecular biology and biotechnology for dealing with global warming: The biosciences will have to play a leading role in developing new technologies for mitigating the impact of greenhouse gas emissions'. *EMBO reports* 17:946–948. https://doi.org/10.15252/embr.201642753

IASC (2016). *Invasive Species Impacts on Infrastructure*. Washington, D.C.

International Genetically Engineered Machine Foundation (iGEM) (2017). *Safety Policies*. Available at: http://2017.igem.org/Safety/Policies (Accessed: 25 July 2018).

International Genetically Engineered Machine (iGEM) (2017). *Registry of Standard Biological Parts*. Available at: http://igem.org/Registry (Accessed: 31 December 2017).

International Genetically Engineered Machine Foundation (iGEM) (2018). *iGEM Previous Competitions*. Available at: http://igem.org/

Previous_Competitions. https://doi.org/10.1142/9789814579551_0008

International Genetically Engineered Machine (iGEM) Foundation (2018). *iGEM Human Practices*. Available at: http://igem.org/Human_Practices (Accessed: 16 July 2018). https://doi.org/10.1142/9789814579551_0008

Ikeda, M., Matsuyama, S., Akagi, S., Ohkoshi, K., Nakamura, S., Minabe, S., Kimura, K. and Hosoe, M. (2017). 'Correction of a Disease Mutation using CRISPR/Cas9-assisted Genome Editing in Japanese Black Cattle'. *Scientific Reports* 7(1):17827. https://doi.org/10.1038/s41598-017-17968-w

Impossible Foods (2018). *Impossible Foods*. Available at: https://impossiblefoods.com/.

International Centre of Insect Physiology and Ecology (2018). *Push-Pull IPM Technology*.

International Civil Society Working Group on Synthetic Biology (2011). *A Submission to the Convention on Biological Diversity's Subsidiary Body on Scientific, Technical and Technological Advice (SBSTTA) on the Potential Impacts of Synthetic Biology on the Conservation and Use of Synthetic Biology*. https://doi.org/10.1002/9780470977873.ch14

Intergovernmental Science Policy Platform on Biodiversity and Ecosystem Services (IPBES) (2016a). *IPBES Assessment Report on Pollinators, Pollination and Food Production: Summary for Policymakers*. https://doi.org/10.3724/sp.j.1003.2012.02144

Intergovernmental Science Policy Platform on Biodiversity and Ecosystem Services (IPBES) (2016b). *Summary for policymakers of the assessment report on the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services on pollinators, pollination and food production*. Bonn. https://doi.org/10.3724/sp.j.1003.2012.02144

International Union for Conservation of Nature (IUCN) (2000). *IUCN Guidelines for the Prevention of Biodiversity Loss Caused by Alien Invasive Species*. Gland, Switzerland: IUCN. https://portals.iucn.org/library/node/12673

International Union for Conservation of Nature (IUCN) (2014). *A quarter of sharks and rays threatened with extinction*. Available at: https://www.iucn.org/content/quarter-sharks-and-rays-threatened-extinction (Accessed: 16 July 2018).

International Union for Conservation of Nature (IUCN) (2016). *The Honolulu Challenge on Invasive Alien Species*. Available at: https://www.iucn.org/theme/species/our-work/invasive-species/honolulu-challenge-invasive-alien-species (Accessed: 29 June 2018). https://doi.org/10.6027/tn2015-517

International Union for Conservation of Nature (IUCN) (2017). *IUCN Red List of Threatened Species Summary Statistics*. Available at: www.iucnredlist.org/about/summary-statistics.

International Union for Conservation of Nature (IUCN) (2018). *The IUCN Red List of Threatened Species. Version 2018-1.* Available at: http://www.iucnredlist.org (Accessed: 1 August 2018).

International Union for Conservation of Nature Species Survival Commission (IUCN SSC) (2016). *IUCN SSC Guiding Principles on Creating Proxies of Extinct Species for Conservation Benefit*. Gland, Switzerland. https://doi.org/10.1603/ice.2016.107949

International Union for Conservation of Nature Species Survival Commission (IUCN SSC) Antelope Specialist Group (2013). *Oryx leucoryx. The IUCN Red List of Threatened Species 2013.* https://doi.org/10.2305/iucn.uk.2011-1.rlts.t15569a4824960.e

International Union for Conservation of Nature (IUCN) World Conservation Congress (2000). *WCC 2000 RES 031 Genetically Modified Organisms and Biodiversity*. Available at: https://portals.iucn.org/library/node/44546 (Accessed: 28 November 2018).

International Union for Conservation of Nature (IUCN) World Conservation Congress (2004). *RES 3.007 A moratorium on the further release of Genetically Modified Organisms (GMOs)*. Available at: https://portals.iucn.org/library/sites/library/files/resrecfiles/WCC_2004_RES_7_EN.pdf (Accessed: 16 July 2018).

International Union for Conservation of Nature (IUCN) World Conservation Congress (2004). *RES 3.008 Genetically Modified Organisms (GMOs) and biodiversity*. Available at: https://portals.iucn.org/library/sites/library/files/resrecfiles/WCC_2004_RES_8_EN.pdf (Accessed: 16 July 2018).

Iqbal, Z., Sattar, M.N. and Shafiq, M. (2016). 'CRISPR/Cas9: a tool to circumscribe cotton leaf curl disease'. *Frontiers in Plant Science* 7:475. https://doi.org/10.3389/fpls.2016.00475

Ishii, T. and Araki, M. (2017). 'A future scenario of the global regulatory landscape regarding genome-edited crops'. *GM Crops and Food*, 8(1):44–56. https://doi.org/10.1080/21645698.2016.1261787

Ito, R., Mustapha, M.M., Tomich, A.D., Callaghan, J.D., McElheny, C.L., Mettus, R.T., Shan, R.M.Q., Sluis-Cremer, N. and Doi, Y. (2017). 'Widespread fosfomycin resistance in Gram-negative bacteria attributable to the chromosomal *fosA* gene'. *mBio* 8(4). https://doi.org/10.1128/mbio.00749-17

Jacobs, D.F., Dalgleish, H.J. and Nelson, C.D. (2013). 'A conceptual framework for restoration of threatened plants: the effective model of American chestnut (Castanea dentata) reintroduction'. *New Phytologist* 197(2):378–393. https://doi.org/10.1111/nph.12020

James, S., Collins, F.H., Welkhoff, P.A., Emerson, C., Godfray, H.C.J., Gottlieb, M., Greenwood, B., Lindsay, S.W., Mbogo, C.M., Okumu, F.O., Quemada, H., Savadogo, M., Singh, J.A., Tountas, K.H. and Toure, Y.T. (2018). 'Pathway to Deployment of Gene Drive Mosquitoes as a Potential Biocontrol Tool for Elimination of Malaria in Sub-Saharan Africa: Recommendations of a Scientific Working Group'. *The American Journal of Tropical Medicine and Hygiene* 98(6 Suppl):1-49. https://doi.org/10.4269/ajtmh.18-0083

Jasanoff, S. (2003). 'Technologies of humility: Citizen participation in governing science'. *Minerva* 41(3):223–244.

Jasny, B.R., Chin, G., Chong, L. and Vignieri, S. (2011). 'Again, and again, and again…'. American Association for the Advancement of Science.

Jaynes, R. (1964). 'Interspecific Crosses in the Genus Castanea'. *Silvae Genetica* 13:146–154.

Jensen, K.K., Gamborg, C., Madsen, K.H., Jorgensen, R.B., von Krauss, M.K., Folker, A.P. and Sandoe, P. (2003). 'Making the EU "Risk Window" transparent: The normative foundations of the environmental risk assessment of GMOs'. *Environmental Biosafety Research* 2(3):161–171. https://doi.org/10.1051/ebr:2003011

Jepson, P.R. and Arakelyan, I. (2017). 'Developing publicly acceptable tree health policy: public perceptions of tree-breeding solutions to ash dieback among interested publics in the UK'. *Forest Policy and Economics* 80:167–177. https://doi.org/10.1016/j.forpol.2017.03.002

Jerez, C.A. (2017). 'Biomining of metals: how to access and exploit natural resource sustainably'. *Microbial biotechnology* 10(5):1191–1193. https://doi.org/10.1111/1751-7915.12792

Jiang, C.-J., Shimono, M., Maeda, S., Inoue, H., Mori, M., Hasegawa, M., Sugano, S. and Takatsuji, H. (2009). 'Suppression of the rice fatty-acid desaturase gene OsSSI2 enhances resistance to blast and leaf blight diseases in rice'. *Molecular Plant-Microbe Interactions* 22(7):820–829. https://doi.org/10.1094/mpmi-22-7-0820

Johnson, J.A., Altwegg, R., Evans, D.M., Ewen, J.G. and Gordon, I.J. (2016). 'Is there a future for genome-editing technologies in conservation?'. *Animal Conservation* 19(2):97–101.

Jones, H.P., Holmes, N.D., Butchart, S.H.M., Tershy, B.R., Kappes, P.J., Corkery, I., Aguirre-Munoz, A., Armstrong, D.P., Bonnaud, E., Burbidge, A.A., Campbell, K., Courchamp, F., Cowan, P.E., Cuthbert, R.J., Ebbert, S., Genovesi, P., Howald, G.R., Keitt, B.S., Kress, S.W., Miskelly, C.M., Oppel, S., Poncet, S., Rauzon, M.J., Rocamora, G., Russell, J.C., Samaniego-Herera, A., Seddon, P.J., Spatz, D.R., Towns, D.R. and Croll, D.A. (2016). 'Invasive mammal eradication on islands results in substantial conservation gains'. *Proceedings of the National Academy of Sciences* 113(15):4033–4038.

Jones, K.R., Venter, O., Fuller, R.A., Allan, J.R., Maxwell, S.L., Negret, P.J. and Watson J.E.M. (2018). 'One-third of global protected land is under intense human pressure'. *Science* 360(6390):788–791.

Joseph, K. and Nithya, N. (2009). 'Material flows in the life cycle of leather'. *Journal of Cleaner Production* 17:676–682. https://doi.org/10.1016/j.jclepro.2008.11.018

Judge, S., Gaudioso, J.M., Hsu, B.H., Camp, R.J. and Hart, P. (2018). *Pacific Island Landbird Monitoring Annual Report, Haleakalā National Park and East Maui Island, 2017. Natural Resource Technical Report NPS/PACN/NRTR—2018/xxx*. Fort Collins, Colorado.

Junges, C. M., Maglianese, M., Lajmanovich, R.C., Peltzer, P.M. and Attademo, A.M., (2017). 'Acute Toxicity and Etho-toxicity of Three Insecticides Used for Mosquito Control on Amphibian Tadpoles'. *Water, Air, & Soil Pollution* 228(4). https://doi.org/10.1007/s11270-017-3324-6

Jurkowski, T.P., Ravichandran, M. and Stepper, P. (2015). 'Synthetic epigenetics—towards intelligent control of epigenetic states and cell identity'. *Clinical epigenetics* 7(1):18. https://doi.org/10.1186/s13148-015-0044-x

KaramiNejadRanjbar, M., Eckermann, K.N., Ahmed, H.M.M., Sanchez, C., H.M., Dippel, S., Marshall, J.M. and Wimmer, E.A. (2018). 'Consequences of resistance evolution in a Cas9-based sex conversion-suppression gene drive for insect pest management'. *Proceedings of the National Academy of Sciences* 115(24):6189–6194. https://doi.org/10.1073/pnas.1713825115.

Kayal, M., Vercelloni, J., de Loma, T.L., Bosserelle, P., Chancerelle, Y., Geoffroy, S., Stievenart, C., Michonneau, F., Penin, L., Planes, S. and Adjeroud, M. (2012). 'Predator crown-of-thorns starfish (*Acanthaster planci*) outbreak, mass mortality of corals, and cascading effects on reef fish and benthic communities'. *PloS one* 7(10):e47363. https://doi.org/10.1371/journal.pone.0047363

Kazana, V., Tsourgiannis, L., Iakovoglou, V., Stamatiou, C., ALexandrov, A., Araujo, S., Bogdan, S., Bozic, G., Brus, R., Bossinger G., Boutsimea, A., Celepirovic, N., Cvrckova, H., Fladung, M., Ivankovic, M., Kazaklis, A., Koutsona, P., Luthar, Z., Machova, P., Mala, J., Mara, K., Mataruga, M., Moravcikova, J., Paffetti, D., Paiva, J.A.P., Raptis, D., Sanchez, C., Sharry, S., Salaj, T., Sijacic/Nikolic, M., Tel Zur, N., Tsvetkov, I., Vettori, C. and Vidal, N. (2015). 'Public attitudes towards the use of transgenic forest trees: a cross-country pilot survey'. *iForest-Biogeosciences and Forestry* 9(2):344.

Kelle, A. (2009). 'Ensuring the security of synthetic biology-towards a 5p governance strategy'. *Systems and Synthetic Biology* 3(1):85–90. https://doi.org/10.1007/s11693-009-9041-8

Kellogg, S.T., Chatterjee, D.K. and Chakrabarty, A.M. (1981). 'Plasmid-assisted molecular breeding: new technique for enhanced biodegradation of persistent toxic chemicals'. *Science* 214(4525):1133–1135. https://doi.org/10.1126/science.7302584

Kelly, J., Sadeghieh, T. and Adeli, K. (2014). 'Peer review in scientific publications: benefits, critiques, & a survival guide'. *EJIFCC* 25(3):227.

Kerr, J. and Landry, J. (2017). *Pulse of the Fashion Industry*.

Kershen, D.L. (2015). 'Sustainability Council of New Zealand Trust v. The Environmental Protection Authority: Gene editing technologies and the law'. *GM Crops & Food* 6(4):216–222. https://doi.org/10.1080/21645698.2015.1122859

Keulartz, J. and van den Belt, H. (2016). 'DIY-Bio–economic, epistemological and ethical implications and ambivalences'. *Life Sciences, Society and Policy* 12(1):7. https://doi.org/10.1186/s40504-016-0039-1

Keung, A.J., Joung, J.K., Khalil, A.S. and Collins, J.J. (2015). 'Chromatin regulation at the frontier of synthetic biology'. *Nature Reviews Genetics* 16(3):159. https://doi.org/10.1038/nrg3900

Killeen, G.F., Tatarsky, A., Diabate, A., Chaccour, C.J., Marshall, J.M., Okumu, F.O., Brunner, S., Newby, G., Williams, Y.A., Malone, D., Tusting, L.S. and Gosling, R.D. (2017). 'Developing an expanded vector control toolbox for malaria elimination'. *BMJ Global Health* 2(2):e000211. https://doi.org/10.1136/bmjgh-2016-000211

Kinchy, A. (2012). *Seeds, science, and struggle: The global politics of transgenic crops*. MIT Press.

Kingiri, A.N. and Hall, A. (2012). 'The Role of Policy Brokers: The Case of Biotechnology in Kenya'. *Review of Policy Research* 29(4):492–522. https://doi.org/10.1111/j.1541-1338.2012.00573.x

Kittinger, J.N., Teneva, L.T., Koike, H., Stamoulis, K.A., Kittinger, D.S., Oleson, K.L.L., Conklin, E., Gomes, M., Wilcox, B. and Friedlander, A.M. (2015). 'From reef to table: social and ecological factors affecting coral reef fisheries, artisanal seafood supply chains, and seafood security'. *PloS one* 10(8). https://doi.org/10.1371/journal.pone.0123856

Klein, R., Tischler, J.S., Muhling, M. and Schlomann, M. (2013). 'Bioremediation of mine water'. In: *Geobiotechnology I*, pp. 109–172. Springer. https://doi.org/10.1007/10_2013_265

Kleinman, D.L. and Vallas, S.P. (2006). 'Contradiction in convergence: Universities and industry in the biotechnology field'. In: *The new political sociology of science: Institutions, networks, and power*, pp. 35–62. The University of Wisconsin Press Madison, WI.

Kleinschmidt, I., Bradley, J., Knox, T.B., Mnzava, A.P., Kafy, H.T., Mbogo, C., Ismail, B.A., Bigoga, J.D., Adechoubou, A., Raghavendra, K., Cook, J., Malik, E.M., Nkuni, Z.J., Macdonald, M., Bayoh, N., Ochomo, E., Fondjo, E., Awono-Ambene, H.P., Etang, J., Akogbeto, M., Bhatt, R.M., Chourasia, M.K., Swain, D.I., Kinyari, T., Subramaniam, K., Massougbodji, A., Oke-Sopoh, M., Ogouyemi-Hounto, A., Kouambeng, C., Abdin, M.S., West, P., Elmardi, K., Cornelie, S., Corbel, V., Valecha, N., Mathenge, E., Kamau, L., Lines, J. and Donnelly, M.J. (2018). 'Implications of insecticide resistance for malaria vector control with long-lasting insecticidal nets: a WHO-coordinated, prospective, international, observational cohort study'. *The Lancet Infectious Diseases* 18(6):640–649. https://doi.org/10.1016/S1473-3099(18)30172-5

Klocko, A.L., Lu, H., Magnuson, A., Brunner, A.M., MA, C. and Strauss, S.H. (2018). 'Phenotypic expression and stability in a large-scale field study of genetically engineered poplars containing sexual containment transgenes'. *Frontiers in Bioengineering and Biotechnology* 6:100. https://doi.org/10.3389/fbioe.2018.00100

Knight, G. M., Dharan, N.J., Fox, G.J., Stennis, N., Zwerling, A., Khurana, R. and Dowdy, D.W. (2016). 'Bridging the gap between evidence and policy for infectious diseases: How models can aid public health decision-making'. *International Journal of Infectious Diseases* 42:17–23. https://doi.org/10.1016/j.ijid.2015.10.024

Knorr-Cetina, K. (1999). *Epistemic cultures: How the sciences make knowledge*. Cambridge, Mass: Harvard University Press.

Kofler, N., Collins, J.P., Kuzma, J., Marris, E., Esvelt, K., Nelson, M.P., Newhouse, A., Rothschild, L.J., Vigliotti, V.S., Semenov, M., Jacobsen, R., Dahlman, J.E., Prince, S., Caccone, A, Brown, T. and Schmitz, O.J. (2018). 'Editing nature: Local roots of global

governance'. *Science* 362(6414):527–529. https://doi.org/10.1126/science.aat4612

Kokotovich, A.E. (2014). 'Delimiting the Study of Risk: Exploring Values and Judgment in Conflicting GMO Ecological Risk Assessment Guidelines'. In: *Contesting Risk: Science, Governance and the Future of Plant Genetic Engineering*, pp. 13–67. Dissertation, University of Minnesota. http://hdl.handle.net/11299/168257

Kolbert, E. (2014). *The Sixth Extinction: An Unnatural History*. New York: Henry Holt and Company.

Kolodziejczyk, B. (2017). *Do-it-yourself biology shows safety risks of an open innovation movement*, *Techtank*. Available at: https://www.brookings.edu/blog/techtank/2017/10/09/do-it-yourself-biology-shows-safety-risks-of-an-open-innovation-movement/.

de Koning, A.P.J., Gu, W., Castoe, T.A., Batzer, M.A. and Pollock, D.D. (2011). 'Repetitive Elements May Comprise Over Two-Thirds of the Human Genome'. *PLoS Genetics* 7(12):e1002384. https://doi.org/10.1371/journal.pgen.1002384

Koplow, D.A. (2004). 'Deliberate Extinction: Whether to Destroy the Last Smallpox Virus'. *Suffolk University Law Review* 37:1–50. https://doi.org/10.1525/sp.2007.54.1.23

Krend, K.L. (2011). 'Avian malaria on Oahu: Disease ecology, population genetics, and the evolution of resistance in Oahu Amakihi'. Dissertation, University of Hawai'i, Manoa.

Krimsky, S. (2004). *Science in the private interest: Has the lure of profits corrupted biomedical research?* Rowman & Littlefield.

Krimsky, S. (2013). 'Do financial conflicts of interest bias research? An inquiry into the "funding effect" hypothesis'. *Science, Technology, & Human Values* 38(4):566–587. https://doi.org/10.1177/0162243912456271

Krisfalusi-Gannon, J., Ali, W., Dellinger, K., Robertson, L., Brady, T.E., Goddard, M.K.M., Tinker-Kulberg, R., Kepley, C.L. and Dellinger, A.L. (2018). 'The Role of Horseshoe Crabs in the Biomedical Industry and Recent Trends Impacting Species Sustainability'. *Frontiers in Marine Science* 5(June):1–13. https://doi.org/10.3389/fmars.2018.00185

Kuhn, T. S. (1970). *The structure of scientific revolutions*, 2nd ed. Chicago: University of Chicago Press.

Kuiken, T. (2016). 'Governance: Learn from DIY biologists'. *Nature* 531/7593):167–168. https://doi.org/10.1038/531167a

Kuiken, T. (2017). *DARPA's Synthetic Biology Initiatives Could Militarize the Environment: Is that something we're comfortable with?*, Slate. Available at: http://www.slate.com/articles/technology/future_tense/2017/05/what_happens_if_darpa_uses_synthetic_biology_to_manipulate_mother_nature.html.

Kumar, S. and Rai, A. (2006). 'Synthetic biology: The intellectual property puzzle'. *Texas Law Review* 85:1745.

Kumar, S. and Rai, A. (2007). 'Synthetic biology: The intellectual property puzzle'. *Texas Law Review* 85:1745.

Kungulovski, G. and Jeltsch, A. (2016). 'Epigenome editing: state of the art, concepts, and perspectives'. *Trends in Genetics* 32(2):101–113. https://doi.org/10.1016/j.tig.2015.12.001

Kupferschmidt, K. (2018). 'Biologists raise alarm over changes to biopiracy rules'. *Science* 361(6397):14–14. https://doi.org/10.1126/science.361.6397.14

Kuzma, J. (2016). 'Reboot the debate on genetic engineering: arguments about whether process or product should be the focus of regulation are stalling progress'. *Nature* 531(7593):165–168. https://doi.org/10.1038/531165a

Kyrou, K., Hammond, A.M., Galizi, R., Kranjc, N., Burt, A., Beaghton, A.K. Nolan, T. and Crisanti, A. (2018). 'A CRISPR–Cas9 gene drive targeting doublesex causes complete population suppression in caged *Anopheles gambiae* mosquitoes'. *Nature biotechnology* 36:1062–1066. https://doi.org/10.1038/nbt.4245

Laird, S.A. and Wynberg, R.P. (2018). *A Fact-Finding and Scoping Study on Digital Sequence Information on Genetic Resources in the Context of the Convention on Biological Diversity and the Nagoya Protocol*. Montreal.

Lan, X., Yao, Z., Zhou, Y., Shang, J., Lin, H., Nuss, D.L. and Chen, B. (2008). 'Deletion of the cpku80 gene in the chestnut blight fungus, *Cryphonectria parasitica*, enhances gene disruption efficiency'. *Current Genetics* 53(1):59–66. https://doi.org/10.1007/s00294-007-0162-x

Lander, E.S. (2015). 'Brave new genome'. *New England Journal of Medicine* 373(1):5–8. https://doi.org/10.1056/NEJMp1506446

Langfeldt, L. (2006). 'The policy challenges of peer review: managing bias, conflict of interests and interdisciplinary assessments'. *Research Evaluation* 15(1):31–41. https://doi.org/10.3152/147154406781776039

LaPointe, D.A., Atkinson, C.T. and Jarvi, S.I. (2009). 'Managing disease'. In: T. K. Pratt, C. T. Atkinson, P. C. Banko, J. D. Jacobi and B. L.

Woodworth (eds.) *Conservation biology of Hawaiian forest birds*, pp. 405–424. New Haven, CT: Yale University Press.

LaPointe, D.A., Atkinson, C.T. and Samuel, M.D. (2012). 'Ecology and conservation biology of avian malaria'. *Annals of the New York Academy of Sciences* 1249(1):211–226. https://doi.org/10.1111/j.1749-6632.2011.06431.x

Lautenbach, S., Seppelt, R., Liebscher, J. and Dormann, C.F. (2012). 'Spatial and Temporal Trends of Global Pollination Benefit'. *PLoS one* 7(4):e35954. https://doi.org/10.1371/journal.pone.0035954 https://doi.org/10.1371/journal.pone.0035954

Lawson, C. and Adhikari, K. (2018). *Biodiversity, Genetic Resources and Intellectual Property*. Routledge. https://doi.org/10.4324/9781315098517

Leduc, S. (1912). *Études de biophysique, 2*. Paris: A. Poinat.

Lee, J.H. and Pijut, P.M. (2017). 'Adventitious shoot regeneration from in vitro leaf explants of *Fraxinus nigra*'. *Plant Cell, Tissue and Organ Culture (PCTOC)* 130(2):335–343. https://doi.org/10.1007/s11240-017-1228-1

Lentzos, F. (2015). 'Synthetic Biology's Defence Dollars: Signals and Perceptions'. *PLOS Synbio Community*. Available at: https://blogs.plos.org/synbio/2015/12/24/synthetic-biologys-defence-dollars-signals-and-perceptions/.

Lentzos, F. (2016). 'Biology's Misuse Potential'. *Connections: The Quarterly Journal* 15(2):48–64.

Levin, R.A., Voolstra, C.R., Agarwal, S., Steinberg, P.D., Suggett, D.J. and van Oppen, M.J.H. (2017). 'Engineering strategies to decode and enhance the genomes of coral symbionts'. *Frontiers in Microbiology* 8:1220. https://doi.org/10.3389/fmicb.2017.01220

Lewin, H.A., Robinson, G.E., Kress, W.J., Baker, W.J., Coddington, J., Crandall, K.A., Durbin, R., Edwards, S.V., Forest, R., Gilbert, T.P., Goldstein, M.M., Grigoriev, I.V., Hackett, K.J., Haussler, D., Jarvis, E.D., Johnson, W.E., Patrinos, A., Richards, S., Castilla-Rubio, J.C., van Sluys, M-A., Soltis, P.S., Yang, H. and Zhang, G. (2018). 'Earth BioGenome Project: Sequencing life for the future of life'. *Proceedings of the National Academy of Sciences* 115(17):4325–4333. https://doi.org/10.1073/pnas.1720115115

Liao, W., Atkinson, C.T., LaPointe, D.A. and Samuel, M.D. (2017). 'Mitigating future avian malaria threats to Hawaiian forest birds from climate change'. *PloS one* 12(1). https://doi.org/10.1371/journal.pone.0168880

Littin, K.E., Mellor, D.J., Warburton, B. and Eason, C.T. (2004). 'Animal welfare and ethical issues relevant to the humane control of vertebrate pests'. *New Zealand Veterinary Journal* 52(1):1–10. https://doi.org/10.1080/00480169.2004.36384

Liu, J., Hull, V., Batistella, M., DeFries, R., Dietz, T., Fu, F., Hertel, T.W., Izaurralde, R.C., Lambin, E.F., Li, S., Martinelli, L.A., McConnell, W.J., Moran, E.F., Naylor, R., Ouyang, Z., Polenske, K.R., Reenberg, A., de Miranda Rocha, G., Simmons, C.S., Verburg, P.H., Vitousek, P.M., Zhang, F. and Zhu, C. (2013). 'Framing sustainability in a telecoupled world'. *Ecology and Society* 18(2). https://doi.org/10.5751/ES-05873-180226

Liu, J., Mooney, H., Hull, V., Davis, S.J., Gaskell, J., Hertel, T., Lubchenco, J., Seto, K.C., Gleick, P., Kremen, C. and Shuxin, L. (2015). 'Systems integration for global sustainability'. *Science* 347(6225):1258832. https://doi.org/10.1126/science.1258832

Liu, W., Ren, J., Zhang, J., Song, X., Liu, S., Chi, X., Chen, Y., Wen, Z. and Li, J. (2017). 'Identification and characterization of a neutralizing monoclonal antibody that provides complete protection against *Yersinia pestis*'. *PloS one* 12(5). https://doi.org/10.1371/journal.pone.0177012

Longino, H.E. (1990). *Science as social knowledge: Values and objectivity in scientific inquiry*. Princeton University Press.

de Lorenzo, V. (2008). 'Systems biology approaches to bioremediation'. *Current Opinion in Biotechnology* 19(6):579–589. https://doi.org/10.1016/j.copbio.2008.10.004

Louv, R. (2008). *Last child in the woods: Saving our children from nature-deficit disorder*. Algonquin books.

Lowe, S., Browne, M. Boudjelas, S. and De Poorter, M. (2000). *100 of the world's worst invasive alien species: a selection from the global invasive species database*. Invasive Species Specialist Group Auckland.

Ludlow, K., Smyth, S.J. and Falck-Zepeda, J. (eds.) (2014). *Socio-Economic Considerations in Biotechnology Regulation*. New York, NY: Springer New York. https://doi.org/10.1007/978-1-4614-9440-9

Lyman, E. and Wold, C. (2013). *A CITES reference manual*.

Macnaghten, P., Owen, R., Stilgoe, J., Wynne, B., Azevedo, A., de Campos, A., Chilvers, J., Dagnino, C.R., di Giulio, G., Frow, E., Garvey, B., Groves, C., Hartley, S., Knobel, M., Kobayaski, E., Lehtonen, M., Lezaun, J., Mello, L., Monteiro, M., Pamplona da Costa, J., Rigolin, C., Fondani, B., Staykova, M., Taddei, R., Till, C., Tyfield, D., Wilford, S. and Velho, L. (2014). 'Responsible

innovation across borders: tensions, paradoxes and possibilities'. *Journal of Responsible Innovation* 1(2):191–199. https://doi.org/10.1080/23299460.2014.922249

Maier, J.A.H., Möhrle, R. and Jeltsch, A. (2017). 'Design of synthetic epigenetic circuits featuring memory effects and reversible switching based on DNA methylation'. *Nature Communications* 8:15336. https://doi.org/10.1038/ncomms15336

Mains, J.W., Breisfoard, C.L., Rose, R.I. and Dobson, S.L. (2016). 'Female adult *Aedes albopictus* suppression by *Wolbachia*-infected male mosquitoes'. *Scientific Reports* 6:33846. https://doi.org/10.1038/srep33846

Maloney, T., Phelan, R. and Simmons, N. (2018). *Saving the horseshoe crab: A synthetic alternative to horseshoe crab blood for endotoxin detection*. *PLoS Biol* 16(10): e2006607. https://doi.org/10.1371/journal.pbio.2006607

Manfredo, M.J., Bruskotter, J.T., Teel, T.L., Fulton, D., Schwartz, S.H., Arlinghaus, R., Oishi, S., Uskul, A.K. Redford, K., Kitayama, S. and Sullivan, L. (2017). 'Why social values cannot be changed for the sake of conservation'. *Conservation Biology* 31(4):772–780. https://doi.org/10.1111/cobi.12855

De March, B.G.E., de Wit, C.A. and Muir, D.C.G. (1998). 'Persistent organic pollutants'. *AMAP Assessment Report: Arctic Pollution Issues*. Arctic Monitoring and Assessment Programme Oslo, Norway, pp. 183–371.

Marchant, G.E. (2003). 'From general policy to legal rule: aspirations and limitations of the precautionary principle'. *Environmental Health Perspectives*. National Institute of Environmental Health Science, 111(14):1799.

Marsden, C.D., Ortega-Del Vecchyo, D., O'Brien, D.P., Taylor, J.F., Ramirez, O., Vila, C., Marques-Bonet, T., Schnabel, R.D., Wayne, R.K. and Lohmueller, K.E. (2016). 'Bottlenecks and selective sweeps during domestication have increased deleterious genetic variation in dogs'. *Proceedings of the National Academy of Sciences* 113(1):152–157. https://doi.org/10.1073/pnas.1512501113

Marshall, A. (2014). 'Drought-tolerant varieties begin global march'. *Nature Biotechnlogy* 32, 308. https://doi.org/10.1038/nbt.2875

Marshall, J. M. (2009). 'The effect of gene drive on containment of transgenic mosquitoes'. *Journal of Theoretical Biology* 258(2):250–265. https://doi.org/10.1016/j.jtbi.2009.01.031

Marshall, J. M. and Hay, B. A. (2012). 'Confinement of gene drive systems to local populations: a comparative analysis'. *Journal of Theoretical Biology* 294:153–171. https://doi.org/10.1016/j.jtbi.2011.10.032

Martel, A., Spitzen-van der Sluijs, A., Blooi, M., Bert, W., Ducatelle, R., Fisher, M.C., Woeltjes, A., Bosman, W., Chiers, K., Bossuyt, F. and Pasmans, F. (2013). '*Batrachochytrium salamandrivorans* sp. nov. causes lethal chytridiomycosis in amphibians'. *Proceedings of the National Academy of Sciences* 110 (38):15325–15329. https://doi.org/10.1073/pnas.1307356110

Martel, A., Blooi, M., Adriaensen, C., Van Rooij, P., Beukema, W., Fisher, M.C., Farrer, R.A., Schmidt, B.R., Tobler, U., Goka, K., Lips, K.R., Muletz, C., Zamudio, K.R., Bosch, J., Lotters, S., Wombwell, E., Garner, T.W.J., Cunningham, A.A., Spitzen-van der Sluijs, A., Salvidio, S., Ducatelle, R., Nishikawa, K., Nguyen. T.T., Kolby, J.E., Van Bocxlaer, I., Bossuyt, F. and Pasmans, F. (2014). 'Recent introduction of a chytrid fungus endangers Western Palearctic salamanders'. *Science* 346(6209):630–631. https://doi.org/10.1126/science.1258268

Martin, G. (2017). 'Citizen Salmon'. *Biocoder* 12:7–11.

Matchett, M.R., Biggins, D.E., Carlson, Powell, B. and Rocke, T. (2010). 'Enzootic plague reduces black-footed ferret (Mustela nigripes) survival in Montana'. *Vector-Borne and Zoonotic Diseases* 10(1):27–35. https://doi.org/10.1089/vbz.2009.0053

Mattick, C.S., Landis, A.E., Allenby, B.R. and Genovese, N.J. (2015). 'Anticipatory life cycle analysis of in vitro biomass cultivation for cultured meat production in the United States'. *Environmental Science & Technology* 49:11941–11949. https://doi.org/10.1021/acs.est.5b01614

Maxwell, S L., Fuller, R.A., Brooks, T.M. and Watson, J.E.M. (2016). 'Biodiversity: The ravages of guns, nets and bulldozers'. *Nature* 536(7615):143–145. https://doi.org/10.1038/536143a

McCartney-Melstad, E., Vu, J.K. and Shaffer, H.B. (2018). 'Genomic Data from an Endangered Amphibian Reveal Unforeseen Consequences of Fragmentation by Roads'. *bioRxiv*, p. 306340. https://doi.org/10.1101/306340

McGowan, C.P., Hines, J.E., Nichols, J.D., Lyons, J.E., Smith, D.R., Kalasz, K.S., Niles, L.J., Dey, A.D., Clark, N.A., Atkinson, P.W., Minton C.D.T. and Kendall, W. (2011). 'Demographic consequences of migratory stopover: linking red knot survival to horseshoe crab spawning abundance'. *Ecosphere* 2(6):1–22. https://doi.org/10.1890/ES11-00106.1

McHughen, A. (2016). 'A critical assessment of regulatory triggers for products of biotechnology: Product vs. process'. *GM Crops & Food*

7(3–4):125–158. https://doi.org/10.1080/21645698.2016.1228516

McKenzie, V.J., Kueneman, J.G. and Harris, R.N. (2018). 'Probiotics as a tool for disease mitigation in wildlife: insights from food production and medicine'. *Annals of the New York Academy of Sciences* 1429(1):18–30. https://doi.org/10.1111/nyas.13617

McMahon, T.A., Sears, B.F., Venesky, M.D., Bessler, S.M., Brown, J.M., Deutsch, K., Halstead, N.T., Lentz, G., Tenouri, N., Young, S., Civitello, D.J., Ortega, N., Fites, J.S., Reinert, L.K., Rollins-Smith, L.A., Raffel, T.R. and Rohr, J.R. (2014). 'Amphibians acquire resistance to live and dead fungus overcoming fungal immunosuppression'. *Nature* 511(7508):224. https://doi.org/10.1038/nature13491

McPhee, D., Pin, A., Kizer, L. and Perlman, L. (2014). 'Deriving Renewable Squalane from Sugarcane'. *Cosmetics & Toiletries magazine* 129(6).

Mead, A. T., Hudson, M. and Chagne, D. (2017). *Maori perspectives and gene editing: A discussion paper*.

Mead, A. T. and Ratuva, S. (2007). *Pacific Genes and Life Patents*. Wellington: UNU Institute of Advanced Studies.

Meadow, M. (2017). *Modern Meadow Reveals the Technologies behind its Biofabricated Leather Materials*. Available at: http://www.modernmeadow.com/press-release/modern-meadow-reveals-technologies-behind-biofabricated-leather-materials/ (Accessed: 16 July 2018).

Meckenstock, R.U., Eisner, M., Griegler, C., Lueders, T., Stumpp, C., Aamand, J., Agathos, S.N., Albrechtsen, H.J., Bastiaens, L., Bjerg, P.L., Boon, N., Dejonghe, W., Huang, W.E., Schmidt, S.I., Smolders, E., Sorensen, S.R., Springael, D. and van Breukelen, B.M. (2015). 'Biodegradation: updating the concepts of control for microbial cleanup in contaminated aquifers'. ACS Publications. https://doi.org/10.1021/acs.est.5b00715

Medina, F.M., Bonnaud, E., Vidal, E., Tershy, B.R., Zavaleta, E.S., Donlan, C.J., Keitt, B.S., Le Corre, M., Horwath, S.V. and Nogales, M. (2011). 'A global review of the impacts of invasive cats on island endangered vertebrates'. *Global Change Biology* 17(11):3503–3510. https://doi.org/10.1111/j.1365-2486.2011.02464.x

Meienberg, F., Sommer, L., Lebrecht, T., Lovera, M., Gonzalez, S., Luig, B., von Bremen, V., Steiner, K., Glauser, M. and Kienle, U. (2015). *The bitter sweet taste of stevia*. Available at: https://www.publiceye.ch/fileadmin/files/documents/Biodiversitaet/BD_STEVIA_REPORT_EN.pdf

Meinshausen, N., Hauser, A., Mooij, J.M., Peters, J., Versteeg, P. and Buhlmann, P. (2016). 'Methods for causal inference from gene perturbation experiments and validation'. *Proceedings of the National Academy of Sciences* 113(27):7361–7368. https://doi.org/10.1073/pnas.1510493113

Meinzen-Dick, R.S. and Pradhan, R. (2002). *Legal pluralism and dynamic property rights*. CGIAR Systemwide Program on Collective Action and Property Rights, International Food Policy Research Institute Washington.

Melillo, J.M., Reilly, J.M., Kicklighter, D.W., Gurgel, A.C., Cronin, T.W., Paltsev, S., Felzer, B.S., Wang, X., Sokolov, A.P. and Schlosser, C.S. (2009). 'Indirect emissions from biofuels: how important?'. *Science* 326(5958):1397–1399. https://doi.org/10.1126/science.1180251

Merton, R.K. (1973). *The sociology of science: Theoretical and empirical investigations*. University of Chicago Press.

Meyer, H. (2011). 'Systemic risks of genetically modified crops: the need for new approaches to risk assessment'. *Environmental Sciences Europe* 23(1):7. https://doi.org/10.1186/2190-4715-23-7

Meyerson, L.A. and Mooney, H. A. (2007). 'Invasive alien species in an era of globalization'. *Frontiers in Ecology and the Environment* 5(4):199–208. https://doi.org/10.1890/1540-9295(2007)5[199:IASIAE]2.0.CO;2

Miao, C., Xiao, L., Hua, K., Zou, C., Zhao, Y., Bressan, R.A. and Zhu, J-K. (2018). 'Mutations in a subfamily of abscisic acid receptor genes promote rice growth and productivity'. *Proceedings of the National Academy of Sciences* 115 (23) 6058–6063. https://doi.org/10.1073/pnas.1804774115

Van Mil, A., Hopkins, H. and Kinsella, S. (2017). *Potential uses for genetic technologies: dialogue and engagement research conducted on behalf of the Royal Society Findings Report*.

Miles, A., Harding, N.J., Botta, G., Clarkson, C.S., Antao, T., Kozak, K., Schrider, D.R., Kern, A.D., Redmond, S., Sharakhov, I., Pearson, R.D., Bergey, C., Fontaine, M.C., Donnelly, M.J., Lawniczak, M.K.N., Kwiatkowski, D.P., Donnelly, M.J., Ayala, D., Besansky, N.J., Burt, A., Caputo, B., della Torre, A., Fontaine, M.C., Godfray, C.J., Hahn, M.W., Kern, A.D., Kwiatkowski, D.P., Lawniczak, M.K.N., Midega, J., Neafsey, D.E., O'Loughlin, S., Pinto, J., Riehle, M.M., Sharakhov, I., Vernick, K.D., Weetman, D., Wilding, C.S., White, B.J., Troco, A.D., Pinto, J., Diabate, A., O'Loughlin, Burt, A., Costantini, C.,Rohatgi, K.R., Besansky, N.J., Elissa,

N., Pinto, J., Coulibaly, B., Riehle, M.M., Vernick, K.D., Pinto, J., Dinis, J., Midega, J., Mbogo, C., Bejon, P., Wilding, C.S., Weetman, D., Mawejje, H.D., Donnelly, M.J., Stalker, J., Rockett, K., Drury, E., Mead, D., Jeffreys, A., Hubbart, C., Rowlands, K., Isaacs, A.T., Jyothi, D., Malangone, C., Vauterin, P., Jeffery, B., Wright, I., Hart, L., Kluczynski, K., Cornelius, V., MacInnis, B., Henrichs, C., Giacomantonio,, R., Kwiatkowski, D.P. (2017). 'Genetic diversity of the African malaria vector *Anopheles gambiae*'. *Nature* 552:96–100. https://doi.org/10.1038/nature24995

Miller, W., Hayes, V.M., Ratan, A., Petersen, D.C., Wittekindt, N.E., Miller, J., Walenz, B., Knight, J., Qi, J., Zhao, F., Wang, Q., Bedoya-Reina, O.C., Katiyar, N., Tomsho, L.P., Kasson, L.M., Hardie, R-A., Woodbridge, P., Tindall, E.A., Bertelsen, M.F., Dixon, D., Pyecroft, S., Helgen, K.M., Lesk, A.M., Pringle, T.H., Patterson, N., Zhang, Y., Kreiss, A., Woods, G.M., Jones, M.E. and Schuster, S.C. (2011). 'Genetic diversity and population structure of the endangered marsupial *Sarcophilus harrisii* (Tasmanian devil)'. *Proceedings of the National Academy of Sciences* 108(30):12348–12353. https://doi.org/10.1073/pnas.1102838108

Milliken, F.J. (1987). 'Three types of perceived uncertainty about the environment: State, effect, and response uncertainty'. *Academy of Management Review* 12(1):133–143. https://doi.org/10.5465/amr.1987.4306502

Minkina, O. and Hunter, C.P. (2017). 'Intergenerational transmission of gene regulatory information in Caenorhabditis elegans'. *Trends in Genetics*. Elsevier.

Mitchell, K., Churcher, T.S., Garner, T.W.J. and Fisher, M.C. (2008). 'Persistence of the emerging pathogen *Batrachochytrium dendrobatidis* outside the amphibian host greatly increases the probability of host extinction'. *Proceedings of the Royal Society of London B: Biological Sciences* 275(1632). https://doi.org/10.1098/rspb.2007.1356

Mizrahi, D.S. and Peters, K.A. (2009). 'Relationships between sandpipers and horseshoe crab in Delaware Bay: A synthesis'. In: *Biology and Conservation of Horseshoe Crabs*, pp.65-87. https://doi.org/10.1007/978-0-387-89959-6_4

Modern Meadow (2017). *Modern Meadow Reveals the Technologies behind its Biofabricated Leather Materials*. Available at: http://www.modernmeadow.com/press-release/modern-meadow-reveals-technologies-behind-biofabricated-leather-materials/ (Accessed: 16 July 2018).

Montpetit, É. (2005). 'A Policy Network Explanation of Biotechnology Policy Differences between the United States and Canada'. *Journal of Public Policy* 25(03):339. https://doi.org/10.1017/S0143814X05000358

Mora, C., Tittensor, D.P., Adl, S., Simpson, A.G.B. and Worm, B. (2011). 'How many species are there on Earth and in the ocean?'. *PLoS Biology* 9(8):e1001127. https://doi.org/10.1371/journal.pbio.1001127

Moran, T., Ries, N.M. and Castle, D. (2009). 'A Cause of Action for Regulatory Negligence? The Regulatory Framework for Genetically Modified Crops in Canada and the Potential for Regulator Liability'. *University of Ottawa Law and Technology Journal* 6(1–2):1–23.

Morand, S., Jittapalapong, S. and Kosoy, M. (2015). 'Rodents as hosts of infectious diseases: Biological and ecological characteristics'. *Vector Borne and Zoonotic Diseases* 15(1):1–2. https://doi.org/10.1089/vbz.2015.15.1.intro

Morgera, E. (2005). 'An Update on the Aarhus Convention and its Continued Global Relevance'. *Review of European Community and International Environmental Law* 14(2):138–147. https://doi.org/10.1111/j.1467-9388.2005.00434.x

Morin, P.J. (1983). 'Predation, competition, and the composition of larval anuran guilds'. *Ecological Monographs* 53(2):119–138. https://doi.org/10.2307/1942491

Moro, D., Byrne, M., Kennedy, M., Campbell, S. and Tizard, M. (2018). 'Identifying knowledge gaps for gene drive research to control invasive animal species: The next CRISPR step'. *Global Ecology and Conservation* 13:e00363. https://doi.org/10.1016/j.gecco.2017.e00363

Moss, R. and Schneider, S. (2000). 'Uncertainties in the IPCC TAR: Recommendations to lead authors for more consistent assessment and reporting'. In: R. Pachauri, T. Taniguchi and K. Tanaka (eds.) *Guidance Papers on the Cross Cutting Issues of the Third Assessment Report of the IPCC*, pp. 33–51. Geneva: World Meteorological Organization.

Muli, E., Patch, H., Frazier, M., Frazier, J., Torto, B., Baumgarten, T., Kilonzo, J., Kimani, J. N., Mumoki, F., Masiga, D., Tumlinson, J. and Grozinger, C. (2014). 'Evaluation of the distribution and impacts of parasites, pathogens, and pesticides on honey bee (*Apis mellifera*) populations in East Africa'. *PLoS one* 9(4). https://doi.org/10.1371/journal.pone.0094459

Muller, M.R. (2015). *Genetic resources as natural information: implications for the Convention on Biological Diversity and Nagoya Protocol*. Routledge. https://doi.org/10.4324/9781315754451

Mulligan, A., Hall, L. and Raphael, E. (2013). 'Peer review in a changing world: An international study measuring the attitudes of researchers'. *Journal of the American Society for Information Science and Technology* 64(1):132–161. https://doi.org/10.1002/

asi.22798

Myers, J.H., Simberloff, D., Kuris, A.M. and Carey, J.R. (2000). 'Eradication revisited: dealing with exotic species'. *Trends in Ecology & Evolution* 15(8):316–320. https://doi.org/10.1016/S0169-5347(00)01914-5

Myhr, A.I. (2010). 'The challenge of scientific uncertainty and disunity in risk assessment and management of GM crops'. *Environmental Values* pp. 7–31. https://doi.org/10.3197/096327110X485365

National Academies of Sciences, Engineering, and Medicine (NASEM) (2016a). *Gene Drives on the Horizon*. Washington, D.C.: National Academies Press. https://doi.org/10.17226/23405

National Academies of Sciences, Engineering, and Medicine (NASEM) (2016b). *Genetically Engineered Crops*. Washington, D.C.: National Academies Press. https://doi.org/10.17226/23395

National Academies of Sciences, Engineering, and Medicine (NASEM) (2017a). *Preparing for Future Products of Biotechnology*. National Academies Press.

National Academies of Sciences, Engineering, and Medicine (NASEM) (2017b). *Preparing for Future Products of Biotechnology*. National Academies Press. https://doi.org/10.17226/24605

National Academies of Sciences, Engineering, and Medicine (NASEM) (2018). *Biodefense in the Age of Synthetic Biology*. Washington, D.C.: National Academies Press. https://doi.org/10.17226/24890

National Research Council (NRC) (1983). *Risk assessment in the federal government: managing the process*. National Academies Press.

National Research Council (NRC) (1986). *Pesticide resistance: strategies and tactics for management*. National Academies Press.

National Research Council (NRC) (1996). *Understanding Risk: Informing Decisions in a Democratic Society*. Edited by P. C. Stern and H. V Fineberg. Washington, D.C.: The National Academies Press. https://doi.org/10.17226/5138

National Research Council (NRC) (2013). *Positioning Synthetic Biology to Meet the Challenges of the 21st Century: Summary Report of a Six Academies Symposium Series*. Washington, D.C.

Neafsey, D.E., Waterhouse, R.M., Abai, M.R., Aganezov, S.S., Alekseyev, M.A., Allen, J.E., Amon, J., Arca, B., Arensburger, P., Artemov, G., Assour, L.A., Basseri, H., Berlin, A., Birren, B.W., Blandin, S.A., Brockman, A.I., Burkot, T.R., Burt, A., Chan, C.S., Chauve, C., Chiu, J.C., Christensen, M., Costantini, C., Davidson, V.L., Deligianni, E., Dottorini, T., Dritsou, V., Gabriel, S.B., Guelbeogo, W.M., Hall, AA.B., Han, M.V., Hlaing, T., Hughes, D.S., Jenkins, A.M., Jiang, X., Jungreis, I., Kakani, E.G., Kamali, M., Kemppainen, P., Kennedy, R.C., Kirmitzoglou, I.K., Koekemoer, L.L., Laban, N., Langridge, N., Lawniczak, M.K., Lirakis, M., Lobo, N.F., Lowy, E., MacCallum, R.M.,Mao, C., Masien, G., Mbogo, C., McCarthy, J., Michel, K., Mitchell, S.N., Moore, W., Murphy, K.A., Naumenko, A.N., Nolan, T., Novoa, E.M., O'Loughlin, S., Oringanje, C., Oshagi, M.A., Pakpour, N., Papathanos, P.A., Peery, A.N., Povelones, M., Prakash, A., Price, D.P., Rajaraman, A., Reimer, L.J., RInker, D.C., Rokas, A., Russell, T.L., Sagnon, N., Sharakhova, M.V., Shea, T., Simao, F.A., Simard, F., Slotman, M.A., Somboon, P., Stegniy, V., Struchiner, C.J., Thomas, G.W., TOjo, M., Topalis, P., Tubio, J.M., Unger, M.F., Vontas, J., Walton, C., Wilding, C.S., Willis, J.H., Wu, Y.C., Yan, G., Zdobnov, E.M., Zhou, X., Catteruccia, F., Christophides, G.K., Collins, F.H., Comman, R.S., Crisanti, A., Donnelly, M.J., Emrich, S.J., Fontaine, M.C., Gelbert, W., Hahn, M.W., Hansen, I.A., Howell, P.I., Kafatos, F.C., Kellis, M., Lawson, D., Louis, C., Luckhart, S., Muskavitch, M.A., RIbeiro, J.M., Riehle, M.A., Sharakhov, I.V., Tu, Z., Zwiebel, L.J. and Besansky, N.J. (2015). 'Highly evolvable malaria vectors: The genomes of 16 *Anopheles* mosquitoes'. *Science* 347(6217):1258522. https://doi.org/10.1126/science.1258522

Needham, M., Howe, G. and Petit, J. (2015). 'Forest health biotechnologies: What are the drivers of public acceptance?'. In: *Forest Health Initiative Annual Meeting.* Washington, D.C.

Nelson, B. (2014). 'Cultural divide'. *Nature* 509(7499):152.

Nelson, B.R., Satyanarayana, B., Moh, J.H.Z., Ikhwanuddin, M., Chatterji, A. and Shaharom, F. (2016). 'The final spawning ground of *Tachypleus gigas* (Müller, 1785) on the east Peninsular Malaysia is at risk: a call for action'. *PeerJ* 4:e2232. https://doi.org/10.7717/peerj.2232

Nelson, K.C., Andow, D.A. and Banker, M.J. (2009). 'Problem formulation and option assessment (PFOA) linking governance and environmental risk assessment for technologies: a methodology for problem analysis of nanotechnologies and genetically engineered organisms'. *The Journal of Law, Medicine & Ethics* 37(4):732–748. https://doi.org/10.1111/j.1748-720X.2009.00444.x

NEPAD (2018). *Gene drives for malaria control and elimination in Africa*. Available at: http://www.nepad.org/resource/gene-drives-malaria-control-and-elimination-africa-1.

Newhouse, A.E., Schrodt, F., Liang, H., Maynard, C.A. and Powell, W.A. (2007). 'Transgenic American elm shows reduced Dutch elm disease symptoms and normal mycorrhizal colonization'. *Plant Cell Reports* 26(7):977–987. https://doi.org/10.1007/s00299-007-0313-z

Newhouse, A.E., Oakes, A.D., Pilkey, H.C., Roden, H.E., Horton, T.R. and Powell, W.A. (2018). 'Transgenic American Chestnuts Do Not Inhibit Germination of Native Seeds or Colonization of Mycorrhizal Fungi'. *Frontiers in Plant Science* 9. https://doi.org/10.3389/fpls.2018.01046

Niissalo, M. A., Leong-Skornickova, J., Webb, E.L. and Khew, G.S. (2018). 'Pedigree analyses and next-generation sequencing reveal critically low regeneration in extremely threatened *Zingiber singapurense* (Zingiberaceae)'. *Botanical Journal of the Linnean Society* 187(2):346–361. https://doi.org/10.1093/botlinnean/boy018

Nijman, V. (2010). 'An overview of international wildlife trade from Southeast Asia'. *Biodiversity and Conservation* 19(4):1101–1114. https://doi.org/10.1007/s10531-009-9758-4

Nilsson, C., Aradottir, A.L., Hagen, D., Halldorsson, G., Hoegh, K., Mitchell, R.J., Raulund-Rasmussen, K., Svavarsdottir, K., Tolvanen, A. and Wilson, S.D. (2016). 'Evaluating the process of ecological restoration'. *Ecology and Society* 21(1). https://doi.org/10.5751/ES-08289-210141

Noble, C., Adlam, B., Church, G.M., K.M. Esvelt and Nowak, M.A. (2018). 'Current CRISPR gene drive systems are likely to be highly invasive in wild populations'. *eLife* 7:e33423. https://doi.org/10.7554/eLife.33423

Nogales, M., Martin, A., Tershy, B.R., Donlan, D.J., Veitch, D., Puerta, N., Wood, B. and Alonso, J. (2004). 'A review of feral cat eradication on islands'. *Conservation Biology* 18(2):310–319. https://doi.org/10.1111/j.1523-1739.2004.00442.x

Novak, B. J., Maloney, T. and Phelan, R. (2018). 'Advancing a New Toolkit for Conservation: From Science to Policy'. *The CRISPR Journal* 1(1):11–15. https://doi.org/10.1089/crispr.2017.0019

NYU Biodesign Challenge (2017). *BEECOSYSTEM*. Available at: http://biodesignchallenge.org/nyu-itp (Accessed: 16 July 2018).

O'Connor, L., Plichart, C., sang, A.C., Breisfoard, C.L., Bossin, H.C. and Dobson, S.L. (2012). 'Open release of male mosquitoes infected with a Wolbachia biopesticide: field performance and infection containment'. *PLoS Neglected Tropical Diseases* 6(11):e1797. https://doi.org/10.1371/journal.pntd.0001797

O'Hanlon, S.J., Rieux, A., Farrer, R.A., Rosa, G.M. Waldman, B., Bataille, A., Kosch, T.A., Murray, K.A., Brankovics, B., Fumagalli, M., Martin, M.D. Wales, N., Alvarado-Rybak, M., Bates, K.A., Berger, L., Boll, S., Brookes, L., Clare, F., Courtois, E.A., Cunningham, A.A., Doherty-Bone, T.M., Ghosh, P., Gower, D.J., Hintz, W.E., Hoglund, J., Jenkinson, T.S., Lin, C-F., Laurila, A., Loyau, A., Martel, A., Meurling, S., Miaud, C., Minting, P., Pasmans, F., Schmeller, D.S., Schmidt, B.R., Shelton, J.M.G., Skerratt, L.F., Smith, F., Soto-Azat, C., Spagnoletti, M., Tessa, G., Toledo, L.F., Valenzuela-Sanchez, A., Verster, R., Voros, J., Webb, R.J., Wierzbicki, C., Wombwell, E., Zamudio, K.R., Aanensen, D.M., James, T.Y., Gilbert, M.T., Weldon, C., Bosch, J., Balloux, F., Garner, T.W.J. and Fisher, M.C. (2018). 'Recent Asian origin of chytrid fungi causing global amphibian declines'. *Science* 360(6389):621 LP-627. Available at: http://science.sciencemag.org/content/360/6389/621.abstract.

Oceana (2008). *Unilever to End Use of Shark Products in Cosmetics*. Available at: oceana.org/en/news-media/press-center/press-releases/%0A.

Odin (2018). *The Odin*. Available at: http://www.the-odin.com/about-us/.

Organisation for Economic C-operation and Development (OECD) (2016). 'OECD Science, Technology and Innovation Outlook 2016'. Paris: OECD Publishing.

Oliver, A. (2018). *Behavioral Economics and the Public Acceptance of Synthetic Biology*, *Hastings Center Report* Available at: https://doi.org/10.1002/hast.819

Ong, S. (2018). 'Singapore bets big on synthetic biology'. *Nature*. https://doi.org/10.1038/d41586-018-04123-2

van Oppen, M.J.H., Oliver, J.K., Putnam, H.M. and Gates, R.D. (2015). 'Building coral reef resilience through assisted evolution'. *Proceedings of the National Academy of Sciences* 112(8):2307–2313. https://doi.org/10.1073/pnas.1422301112

van Oppen, M.J.H., Gates, R.D., Blackall, L.L., Chakravarti, L.J., Chan, W.Y., Cormick, C., Crean, A., Damjanovic, K., Epstein, H., Harrison, P.L., Jones, T.A., Miller, M., Pears, R.J., Peplow, L.M., Raftos, D.A., Schaffelke, B., Stewart, K., Torda, G., Wachenfeld, D., Weeks, A.R. and Putnam, H.M. (2017). 'Shifting paradigms in restoration of the world's coral reefs'. *Global Change Biology* 23(9):3437–3448. https://doi.org/10.1111/gcb.13647

Oye, K.A., Esvelt, K., Appleton, E., Catteruccia, F., Church, G., Kuiken, T., Lightfoot, S. B-Y., McNamara, J., Smidler, A. and Collins, J.P.

(2014). 'Regulating gene drives'. *Science* 345(6197):626–628. https://doi.org/10.1126/science.1254287

Pacheco, F. A. L., Fernandes, L.F.S., Valle Junior, R.F., Valera, C.A. and Pissarra, T.C.T. (2018). 'Land degradation: multiple environmental consequences and routes to neutrality'. *Current Opinion in Environmental Science & Health* 5:79–86. https://doi.org/10.1016/j.coesh.2018.07.002

Paillet, F. L. (1993). 'Growth Form and Life Histories of American Chestnut and Allegheny and Ozark Chinquapin at Various North American Sites'. *Bulletin of the Torrey Botanical Club*, 120(3):257–268. https://doi.org/10.2307/2996990

Paillet, F. L. (2002). 'Chestnut: history and ecology of a transformed species'. *Journal of Biogeography* 29(10-11):1517–1530. https://doi.org/10.1046/j.1365-2699.2002.00767.x

Palla, K. J. and Pijut, P. M. (2015). 'Agrobacterium-mediated genetic transformation of *Fraxinus americana* hypocotyls'. *Plant Cell, Tissue and Organ Culture (PCTOC)* 120(2):631–641. https://doi.org/10.1007/s11240-014-0630-1

Paoletti, C., Flamm, E., Yan, W., Meek, S., Renckens, S., Fellous, M. and Kuiper, H. (2008). 'GMO risk assessment around the world: Some examples'. *Trends in Food Science and Technology* 19(SUPPL. 1):70–78. https://doi.org/10.1016/j.tifs.2008.07.007

Parkes, J., Fisher, P., Robinson, S. and Aguirre-Munoz, A., (2014). 'Eradication of feral cats from large islands: an assessment of the effort required for success'. *New Zealand Journal of Ecology* pp. 307–314.

Parks, S., Ghiga, I., Lepetit, L., Parris, S., Chataway, J. and Jones, M.M. (2017). 'Developing standards to support the synthetic biology value chain'. RAND Corporation. https://doi.org/10.7249/RR1527

Parrish, C.C. (2009). 'Essential fatty acids in aquatic food webs'. In: *Lipids in Aquatic Ecosystems*, pp. 309–326. Springer. https://doi.org/10.1007/978-0-387-89366-2_13

Pauwels, K., Mampuys, R., Golstein, C., Breyer, D., Herman, P., Kaspari, M., Pages, J-C., Pfister, H., van der Wilk, F., Schonig, B. (2013). 'Event report: SynBio Workshop (Paris 2012) – Risk assessment challenges of Synthetic Biology'. *Journal für Verbraucherschutz und Lebensmittelsicherheit* 8(3):215–226. https://doi.org/10.1007/s00003-013-0829-9

Paxton, E.H., Camp, R.J., Gorrensen, P.M., Crampton, L.H., Leonard Jr, D.L. and VanderWerf, E.A. (2016). 'Collapsing avian community on a Hawaiian island'. *Science Advances* 2(9):e1600029. https://doi.org/10.1126/sciadv.1600029

Paxton, E.H., Gorresen, P. M. and Camp, R. J. (2013). *Abundance, distribution, and population trends of the iconic Hawaiian Honeycreeper, the 'I'iwi (*Vestiaria coccinea*) throughout the Hawaiian Islands*. U.S. Geological Survey Open-File Report 2013-1150, 59 p. https://doi.org/10.3133/ofr20131150

Pei, L., Gaisser, S. and Schmidt, M. (2012). 'Synthetic biology in the view of European public funding organisations'. *Public Understanding of Science* 21(2):149–162. https://doi.org/10.1177/0963662510393624

Pejchar, L. and Mooney, H.A. (2009). 'Invasive species, ecosystem services and human well-being'. *Trends in Ecology & Evolution* 24(9):497–504. https://doi.org/10.1016/j.tree.2009.03.016

Personal Care Council (2003). *CIR Ingredient Status Report*. Available at: https://online.personalcarecouncil.org/jsp/CIRList.jsp?id=591 (Accessed: 16 July 2018).

Peterson, D. C. (2006). 'Precaution: principles and practice in Australian environmental and natural resource management'. *Australian Journal of Agricultural and Resource Economics* 50(4):469–489. https://doi.org/10.1111/j.1467-8489.2006.00372.x

Piaggio, A.J., Segelbacher, G., Seddon, P.J., Alphey, L., Bennett, E.L., Carlson, R.H., Friedman, R.M., Kanavy, D., Phelan, R., Redford, K.H., Rosales, M., Slobodian, L. and Wheeler, K. (2017). 'Is it time for synthetic biodiversity conservation?'. *Trends in Ecology & Evolution* 32(2):97–107. https://doi.org/10.1016/j.tree.2016.10.016

Pielke Jr, R.A. (2007). *The honest broker: making sense of science in policy and politics*. Cambridge University Press. https://doi.org/10.1017/CBO9780511818110

Pikaar, I., Matassa, S., Bodirsky, B.L., Weindl, I., Humpenoder, F., Rabaey, K, Boon, N., Bruschi, M., Yuan, Z., van Zanten, H., Herrero, M., Verstraete, W. and Popp, A. (2018). 'Decoupling Livestock from Land Use through Industrial Feed Production Pathways'. *Environmental Science & Technology*. ACS Publications. https://doi.org/10.1021/acs.est.8b00216

Pimm, S. L., Jenkins, C.N., Abell, R., Brooks, T.M., Gittleman, J.L., Joppa, L.N. Raven, P.H., Roberts, C.M. and Sexton, J.O. (2014). 'The biodiversity of species and their rates of extinction, distribution, and protection'. *Science* 344(6187):1246752. https://doi.org/10.1126/science.1246752

Pisa, L., Goulson, D., Yang, E-C., Gibbons, D., Sanchez-Bayo, F., Mitchell, E., Aebi, A. van der Sluijs, J., MacQuarrie, C.J.K., Giorio, C.,

Long, E.Y., McField, M., van Lexmond, M.B. and Bonmatin, J-M. (2017). 'An update of the Worldwide Integrated Assessment (WIA) on systemic insecticides. Part 2: impacts on organisms and ecosystems'. *Environmental Science and Pollution Research*. Springer Berlin Heidelberg, pp. 1–49. https://doi.org/10.1007/s11356-017-0341-3

Pitt, W.C., Berentsen, A.R.E., Shiels, A.B., Volker, S.F., Eisenmann, J.D., Wegmann, A.S. and Howald, G.R. (2015). 'Non-target species mortality and the measurement of brodifacoum rodenticide residues after a rat (*Rattus rattus*) eradication on Palmyra Atoll, tropical Pacific'. *Biological Conservation* 185:36–46. https://doi.org/10.1016/j.biocon.2015.01.008

Popper, K. (2005). *The logic of scientific discovery*. Routledge. https://doi.org/10.4324/9780203994627

Poppick, L. (2018). *Threatened Species? Science to the (Genetic) Rescue!*, Smithsonian. Available at: https://www.smithsonianmag.com/science-nature/threatened-species-science-genetic-rescue-180963040/ (Accessed: 6 June 2018).

Pottage, A. (2006). 'Too much ownership: Bio-prospecting in the age of synthetic biology'. *BioSocieties* 1(2):137–158. https://doi.org/10.1017/S1745855206050241

Pottage, A. and Marris, C. (2012). 'The cut that makes a part'. *BioSocieties* 7(2):103–114. https://doi.org/10.1057/biosoc.2012.1

Prabhu, K.V (2009). 'Chapter 4: Use of GMOs under containment, confined and limited field trials and post-release monitoring of GMOs'. *Biosafety of Genetically Modified Organisms: Basic concepts, methods and issues*, pp. 157–220.

Pratchett, M.S., Caballes, C.F., Rivera-Posada, J. and Sweatman, H.P.A. (2014). 'Limits to understanding and managing outbreaks of crown-of-thorns starfish (Acanthaster spp.)'. *Oceanography and Marine Biology: An Annual Review*, 52:133–200. https://doi.org/10.1201/b17143-4

Pugh, J. (2016). 'Driven to extinction? The ethics of eradicating mosquitoes with gene-drive technologies'. *Journal of Medical Ethics* 42(9):578–581. https://doi.org/10.1136/medethics-2016-103462

Pyle, R. L. and Pyle, P. (2017). 'The Birds of the Hawaiian Islands: Occurrence, History, Distribution, and Status'. *BP Bishop Museum, Honolulu, HI, USA Version 2*. Available at: http://hbs.bishopmuseum.org/birds/rlp-monograph.

Quadrana, L. and Colot, V. (2016). 'Plant transgenerational epigenetics'. *Annual Review of Genetics* 50:467–491. https://doi.org/10.1146/annurev-genet-120215-035254

Rademeyer, J. (2012). *Killing for profit: Exposing the illegal rhino horn trade*. Penguin Random House South Africa.

Raftery, A. E., Zimmer, A., Frierson, D.M.W., Startz, R. and Liu, P. (2017). 'Less than 2 C warming by 2100 unlikely'. *Nature Climate Change* 7:637–641. https://doi.org/10.1038/nclimate3352

Rai, A. and Boyle, J. (2007). 'Synthetic biology: caught between property rights, the public domain, and the commons'. *PLoS Biology* 5(3):e58. https://doi.org/10.1371/journal.pbio.0050058

Räikkönen, J., Vucetich, J.A., Peterson, R.O. and Nelson, M.P. (2009). 'Congenital bone deformities and the inbred wolves (*Canis lupus*) of Isle Royale'. *Biological Conservation* 142(5):1025–1031. https://doi.org/10.1016/j.biocon.2009.01.014

Rask, M. and Worthington, R. (2015). *Governing Biodiversity Through Democratic Deliberation*. Routledge. https://doi.org/10.4324/9781315849317

Rebollar, E.A., Antwis, R.E., Becker, M.H., Belden, L.K., Bletz, M.C., Brucker, R.M., Harrison, X.A., Hughey, M.C., Kueneman, J.G., Loudon, A.H., McKenzie, V., Medina, D., Minbiole, K.P.C., Rollins-Smith, L.A., Walke, J.B., Weiss, S., Woodhams, D.C. and Harris, R.N. (2016). 'Using "omics" and integrated multi-omics approaches to guide probiotic selection to mitigate chytridiomycosis and other emerging infectious diseases'. *Frontiers in Microbiology* 7:68. https://doi.org/10.3389/fmicb.2016.00068

Redford, K.H., Adams, W., Mace, G., Carlson, R., Sanderson, S. and Aldrich, S. (2013). *How will synthetic biology and conservation shape the future of nature?* A framing paper prepared for a meeting between synthetic biology and conservation professionals, Clare College, Cambridge, UK, 9-12 April 2013. https://secure3.convio.net/wcs/pdf/Synthetic_Biology_and_Conservation_Framing_Paper.pdf

Redford, K.H., Adams, W., Carlson, R. and Mace, G.M. (2014). 'Synthetic biology and the conservation of biodiversity'. *Oryx*. Cambridge University Press, 48(3):330–336. https://doi.org/10.1017/S0030605314000040

Redford, K.H., Adams, W. and Mace, G. M. (2013). 'Synthetic biology and conservation of nature: wicked problems and wicked solutions'. *PLoS Biology* 11(4):e1001530. https://doi.org/10.1371/journal.pbio.1001530

Reed, J.M., DesRochers, D.W., VanderWerf, E.A. and Scott, J.M. (2012). 'Long-term persistence of Hawaii's endangered avifauna

through conservation-reliant management'. *BioScience* 62(10):881–892. https://doi.org/10.1525/bio.2012.62.10.8

Reeves, R.G., Voeneky, S., Caetano-Anollés, D., Beck, F. and Boëte, C. (2018). 'Agricultural research, or a new bioweapon system?' *Science* 362(6410):35–37. https://doi.org/10.1126/science.aat7664

Resnik, D.B. (2018). 'Ethics of community engagement in field trials of genetically modified mosquitoes'. *Developing world bioethics* 18(2):135–143. https://doi.org/10.1111/dewb.12147

Reuss, M. and Cutcliffe, S.H. (2010). *The illusory boundary: Environment and technology in history*. University of Virginia Press.

Revive & Restore (2015). *New Genomic Solutions for Conservation Problems Workshop*. Available at: http://reviverestore.org/meeting-report/ (Accessed: 16 July 2018).

Revive & Restore (2018). *Passenger Pigeon De-extinction*. Available at: http://reviverestore.org/passenger-pigeon-de-extinction/ (Accessed: 1 July 2018).

Revive & Restore, San Diego Zoo Global and Intrexon (2016). 'A genetic rescue proposal for the black-footed ferret: addressing genetic diversity erosion and enhancing disease resistance in the endangered black-footed ferret with a new form of genetic rescue – employing interspecies somatic cell nuclear transfer an', p. 20.

Ricciardi, A., Blackburn, T.M., Carlton, J.T., Dick, J.T.A., Hulme, P.E., Iacarella, J.C., Jeschke, J.M., Liebhold, A.M., Lockwood, J.L., MacIsaac, H.J., Pysek, P., Richardson, D.M., Ruiz, G.M., Simberloff, D., Sutherland, W.J., Wardle, D.A. and Aldridge, D.C. (2017). 'Invasion science: a horizon scan of emerging challenges and opportunities'. *Trends in Ecology & Evolution* 32(6):464–474. https://doi.org/10.1016/j.tree.2017.03.007

Rimmer, M. (2008). *Intellectual property and biotechnology: biological inventions*. Edward Elgar Publishing. https://doi.org/10.4337/9781848440180

Ripple, W.J., Chapron, G., Lopez-Bao, J.V., Durant, S.M., MacDonald, D.W., Lindsey, P.A., Bruskotter, J.T., Campos-Arceiz, A., Corlett, R.T., Darimont, C.T., Dickman, A.J., Dirzo, R., Dublin, H.T., Estes, J.A., Everatt, K.T., Galetti, M., Goswami, V.R., Hayward, M.W., Hedges, S., Hoffman, M., Hunter, L.T.B., Kerley, G.I.H., Letnic, M., Levi, T., Maisels, F., Morrison, J.C., Nelson, M.P., Newsome, T.M., Painter, L., Pringle, R.M., Sandom, C. J., Terborgh, J., Treves, A., Van Valkenburgh, B., Vucetic, J.A., Wirsing, A.J., Wallach, A.D., Wolf, C., Woodroffe, R. Young, H., Zhang, L. (2016). 'Saving the world's terrestrial megafauna'. *Bioscience* 66(10):807–812. https://doi.org/10.1093/biosci/biw092

Rittel, H.W J. and Webber, M.M. (1973). 'Dilemmas in a general theory of planning'. *Policy Sciences* 4(2):155–169. https://doi.org/10.1007/BF01405730

Ritterson, R. (2012). *A Call for a Public, Democratically Deliberative Facet in Synthetic Biology Policymaking*, *LEAP*. Available at: https://www.synbioleap.org/2012-strategic-action-plans-blog/2017/6/8/a-call-for-a-public-democratically-deliberative-facet-in-synthetic-biology-policymaking.

Roberts, A., Andrade, P.P., Okumu, F., Quemada, H., Savadogo, M., Singh, J.A. and James, S. (2017). 'Results from the workshop "problem formulation for the use of gene drive in mosquitoes"'. *American Journal of Tropical Medicine and Hygiene* 96(3):530–533. https://doi.org/10.4269/ajtmh.16-0726

Robin, M.M. (2010). *The World According to Monsanto*. The New Press.

Rodgers, C.P. (2003). 'Liability for the Release of GMOs into the Environment: Exploring the Boundaries of Nuisance'. *Cambridge Law Journal* 62(2):371–402. https://doi.org/10.1017/S0008197303006354

Rodriguez-Escamilla, Z., Martínez-Núñez, M.A. and Merino, E. (2016). 'Epigenetics knocks on synthetic biology's door'. *FEMS Microbiology Letters* 363(17) p. fnw191. https://doi.org/10.1093/femsle/fnw191

Rodríguez-Leal, D., Lemmon, Z.H., Man, J., Bartlett, M.E. and Lippmann, Z.B. (2017). 'Engineering quantitative trait variation for crop improvement by genome editing'. *Cell* 171(2), pp. 470–480. https://doi.org/10.1016/j.cell.2017.08.030

Roelle, J.E. (2006). 'Recovery of the Black-footed Ferret: Progress and Continuing Challenges'. Presented at the Symposium on the Status of the Black-footed Ferret and Its Habitat. Fort Collins, Colorado: US Department of the Interior, US Geological Survey.

Rowe, G. and Frewer, L.J. (2005). 'A typology of public engagement mechanisms'. *Science, Technology, & Human Values* 30(2), pp. 251–290. https://doi.org/10.1177/0162243904271724

Rusche, L.N., Kirchmaier, A.L. and Rine, J. (2003). 'The establishment, inheritance, and function of silenced chromatin in Saccharomyces cerevisiae'. *Annual Review of Biochemistry* 72(1), pp. 481–516. https://doi.org/10.1146/annurev.biochem.72.121801.161547

Russell, J.C. and Holmes, N.D. (2015). 'Tropical island conservation: rat eradication for species recovery'. *Biological Conservation* 185, pp. 1–7. https://doi.org/10.1016/j.biocon.2015.01.009

Russell, J.C., Towns, D.R. and Clout, M.N. (2008). 'Review of rat invasion biology: implications for island biosecurity'. *Science for Conservation* (286). Department of Conservation.

Saegusa, A. (1999). 'Japan tightens rules on GM crops to protect the environment'. Nature Publishing Group. https://doi.org/10.1038/21483

Sala, O.E., Chapin III, F.S., Armesto, J.J., Berlow, E., Bloomfield, J., Dirzo, R., Huber-Sanwald, R., Huenneke, L.F., Jackson, R.B., Kinzig, A., Leemans, R., Lodge, D.M., Mooney, H.A., Oesterheld, M., Poff, N.L., Sykes, M.T., Walker, B.H., Walker, M. and Wall, D.H. (2000). 'Global biodiversity scenarios for the year 2100'. *Science* 287(5459), pp. 1770–1774. https://doi.org/10.1126/science.287.5459.1770

Sanders, J.W. and Ponzio, T.A. (2017). 'Vectored immunoprophylaxis: an emerging adjunct to traditional vaccination'. *Tropical Diseases, Travel Medicine and Vaccines* 3(1):3. https://doi.org/10.1186/s40794-017-0046-0

Sandler, R.L. (2012). *The Ethics of Species: An Introduction*. Cambridge University Press. https://doi.org/10.1017/CBO9781139151221

Sarewitz, D. (1996). *Frontiers of illusion: Science, technology, and the politics of progress*. Temple University Press.

Sarewitz, D. (2015). 'CRISPR: Science can't solve it'. *Nature News*, 522(7557):413. https://doi.org/10.1038/522413a

Sauter, A., Albrecht, S., van Doren, D., Konig, H., Reiss, T. and Trojok, R. (2015). *Synthetic Biology – the next phase of biotechnology and genetic engineering.* Berlin.

Scalera, R., Genovesi, P., Essl, F. and Rabitsch, W. (2012). 'The impacts of invasive alien species in Europe'. *European Environment Agency Technical Report* 16:114.

Schloegel, L.M., Toledo, L.F., Longcore, J.E., Greenspan, S.E., Vieira, C.A., Lee, M., Zhao, S., Wangen, C., Ferreira, C.M., Hipolito, M., Davies, A.J., Cuomo, C.A., Daszak, P. and James, J.Y. (2012). 'Novel, panzootic and hybrid genotypes of amphibian chytridiomycosis associated with the bullfrog trade'. *Molecular Ecology* 21(21):5162–5177. https://doi.org/10.1111/j.1365-294X.2012.05710.x

Schmidt, M., Ganguli-Mitra, A., Torgersen, H., Kelle, A., Deplazes, A. and Biller-Andorno, N. (2009). 'A priority paper for the societal and ethical aspects of synthetic biology'. *Systems and Synthetic Biology* 3(1–4):3–7. https://doi.org/10.1007/s11693-009-9034-7

Schmidt, M.W.I., Torn, M.S., Abiven, S., Dittmar, T., Guggenberger, G., Janssens, I.A., Kleber, M., Kogel-Knabner, I., Lehmann, J., Manning, D.A., Nannipieri, P., Rasse, D.P., Weiner, S. and Trumbore, S.E. (2011). 'Persistence of soil organic matter as an ecosystem property'. *Nature* 478(7367):49. https://doi.org/10.1038/nature10386

Schreiber, E.A. and Burger, J. (2001). *Biology of Marine Birds*. CRC Press. https://doi.org/10.1201/9781420036305

Schurman, R.A., Kelso, D.T. and Kelso, D.D. (2003). *Engineering trouble: Biotechnology and its discontents*. University of California Press.

Schwab, K. (2016). *The Fourth Industrial Revolution: what it means, how to respond*. World Economic Forum 2016. Available at: https://www.weforum.org/agenda/2016/01/the-fourth-industrial-revolution-what-it-means-and-how-to-respond/.

Science for Environment Policy (2016). *Synthetic biology and biodiversity. Future Brief 15.* Bristol. Available at: http://ec.europa.eu/science-environment-policy.

Scientific Committee on Emerging and Newly Identified Health Risks (SCENIHR), Scientific Committee on Consumer Safetey (SCCS) (2014). *Synthetic Biology I Definition, Opinion, 25*.

Sclove, R.E. (2010). 'Reinventing technology assessment'. *Issues in Science and Technology* 27(1):34–38.

Scott, D., Abdelhakim, D., Miranda, M., Hoft, R. and Cooper, H.D. (2015). *Synthetic Biology - CBD Technical Series No. 82*. Montreal.

Secretariat of the Convention on Biological Diversity (2015). *COP 12 Decision XII/24 - New and emerging issues: synthetic biology*. Available at: https://www.cbd.int/decision/cop/default.shtml?id=13387 (Accessed: 25 June 2018).

Servick, K. (2018). 'US lawmakers float plan to regulate cultured meat'. American Association for the Advancement of Science.

Seyfried, G., Pei, L. and Schmidt, M. (2014). 'European Do-it-yourself (DIY) Biology: beyond the hope, hype and horror'. *Bioessays* 36(6):548–551. https://doi.org/10.1002/bies.201300149

Shafer, A.B.A., Wolf, J.B., Alves, P.C., Bergstrom, L., Bruford, M.W., Brannstrom, I., Colling, G., Dalen, L., De Meester, L., Ekblom, R.,

Fawcett, K.D., Fior, S., Hajibabaei, M., Hill, J.A., Hoezel, A.R., Hoglund, J., Jensen, E.L., Krause, J., Kristensen, T.N., Krutzen, M., McKay, J.K., Norman, A.J., Ogden, R., Osterling, E.M., Ouborg, N.J., Piccolo, J., Popovic, D., Primmer, C.R., Reed, F.A., Foumet, M., Salmona, J., Schenekar, T., Schwartz, M.K., Segelbacher, G., Senn, H., Thaulow, J., Valtonen, M., Veale, A., Vergeer, P., Vijay, N., Vila, C., Weissensteiner, M., Wennerstrom, L., Wheat, C.W. and Zielinski, P. (2015). 'Genomics and the challenging translation into conservation practice'. *Trends in Ecology & Evolution* 30(2):78–87. https://doi.org/10.1016/j.tree.2014.11.009

Shapira, P., Kwon, S. and Youtie, J. (2017). 'Tracking the emergence of synthetic biology'. *Scientometrics*. Springer, 112(3), pp. 1439–1469. https://doi.org/10.1007/s11192-017-2452-5

Shapiro, B. (2015). *How to clone a mammoth: the science of de-extinction*. Princeton University Press.

Sharakhov, I.V. and Sharakhova, M.V. (2015). 'Heterochromatin, histone modifications, and nuclear architecture in disease vectors'. *Current Opinion in Insect Science* 10:110–117. https://doi.org/10.1016/j.cois.2015.05.003

Shelton Lab (2018). *Diamond Back Moth Information and Resources*.

Shi, J., Gao, H., Wang, H., Lafitte, H.R., Archibald, R.L., Yang, M., Hakimi, S.M., Mo, H. and Habben, J.E. (2017). 'ARGOS 8 variants generated by CRISPR-Cas9 improve maize grain yield under field drought stress conditions'. *Plant Biotechnology Journal* 15(2):207–216. https://doi.org/10.1111/pbi.12603

Shukla-Jones, A., Friedrichs, S. and Winickoff, D. E. (2018). *Gene editing in an international context: Scientific, economic and social issues across sectors*. OECD Publishing.

Siegrist, M., Sutterlin, B. and Hartmann, C. (2018). 'Perceived naturalness and evoked disgust influence acceptance of cultured meat'. *Meat Science* 139:213–219. https://doi.org/10.1016/j.meatsci.2018.02.007

Simon, S., Otto, M. and Engelhard, M. (2018). 'Synthetic gene drive: between continuity and novelty'. *EMBO reports* 19(5):e45760. https://doi.org/10.15252/embr.201845760

Sinkins, S.P. and Gould, F. (2006). 'Gene drive systems for insect disease vectors'. *Nature Reviews Genetics* 7(6):427. https://doi.org/10.1038/nrg1870

Sismondo, S. (2010). *An introduction to science and technology studies*. Wiley-Blackwell Chichester.

Skvortsova, K., Iovino, N. and Bogdanović, O. (2018). 'Functions and mechanisms of epigenetic inheritance in animals'. *Nature Reviews Molecular Cell Biology*, p. 1. https://doi.org/10.1038/s41580-018-0074-2

Smith, D.R., Brockmann, H.J., Beekey, M.A., King, T.L., Millard, M.J. and Zaldivar-Rae, J. (2016). 'Limulus polyphemus, The American Horseshoe Crab'. In: *The IUCN Red List of Threatened Species*.

Smith, K. (2013). 'Synthetic Biology: A Utilitarian Perspective'. *Bioethics* 27(8):453–463. https://doi.org/10.1111/bioe.12050

Snow, R.W., Sartorius, B., Kyalo, D., Maina, J., Amratia, P., Mundia, C. W., Bejon, P. and Noor, A. M. (2017). 'The prevalence of *Plasmodium falciparum* in sub-Saharan Africa since 1900'. *Nature* 550(7677):515–518. https://doi.org/10.1038/nature24059

Sodhi, N.S., Butler, R., Laurance, W.F. and Gibson, L. (2011). 'Conservation successes at micro-, meso-and macroscales'. *Trends in Ecology & Evolution* 26(11):585–594. https://doi.org/10.1016/j.tree.2011.07.002

SoundBioLab (2018). *Citizen Salmon Project is Making Waves*. Available at: https://sound.bio/soundblog/2018/6/5/citizen-salmon-project-is-making-waves.

Spalding, M., Burke, L., Wood, S.A., Ashpole, J., Hutchison, J. and zu Ermgassen, P.S.E. (2017). 'Mapping the global value and distribution of coral reef tourism'. *Marine Policy* 82:104–113. https://doi.org/10.1016/j.marpol.2017.05.014

Spatz, D.R., Zilliacus, K.M., Holmes, N.D., Butchart, S.H.M., Genovesi, P., Ceballos, G., Tershy, B.R. and Croll, D.A. (2017). 'Globally threatened vertebrates on islands with invasive species'. *Science Advances* 3(10):e1603080. https://doi.org/10.1126/sciadv.1603080

Sprague, M., Betancor, M. B. and Tocher, D. R. (2017). 'Microbial and genetically engineered oils as replacements for fish oil in aquaculture feeds'. *Biotechnology Letters* 39(11):1599–1609. https://doi.org/10.1007/s10529-017-2402-6

St Clair, J. J. H. (2011). 'The impacts of invasive rodents on island invertebrates'. *Biological Conservation* 144(1):68–81. https://doi.org/10.1016/j.biocon.2010.10.006

Stafford, W.H.L., Lotter, G.A., von Maltitz, G.P. and Brent, A.C. (2018). 'Biofuels technology development in Southern Africa'.

Development Southern Africa, pp. 1–20. https://doi.org/10.1080/0376835X.2018.1481732

Stegen, G., Pasmans, F., Schmidt, B.R., Rouffaer, L.O., Van Praet, S., Schaub, M., Canessa, S., Laudelout, A., Kinet, T., Adriaensen, C., Haesebrouck, F., Bert, W., Bossuyt, F. and Martel, A. (2017). 'Drivers of salamander extirpation mediated by *Batrachochytrium salamandrivorans*'. *Nature* 544(7650):353. https://doi.org/10.1038/nature22059

Steiner, K. C., Westbrook, J.W., Hebard, F.V., Georgi, L.L., Powell, W.A. and Fitzsimmons, S.F. (2017). 'Rescue of American chestnut with extraspecific genes following its destruction by a naturalized pathogen'. *New Forests* 48(2):317–336. https://doi.org/10.1007/s11056-016-9561-5

Stemerding, D., Rerimassie, V., Srinivas, R. and Zhang, W. (2014). *Ethics Debates on Synthetic Biology in the Three Regions*.

De Steur, H., Mehta, S., Gellynck, X. and Finkelstein, J.L. (2017). 'GM biofortified crops: potential effects on targeting the micronutrient intake gap in human populations'. *Current Opinion in Biotechnology* 44:181–188. https://doi.org/10.1016/j.copbio.2017.02.003

Stewart, R.B. (2003). 'Administrative Law in the Twenty-First Century'. *New York University Law Review*, 78, pp. 437–51.

Stilgoe, J., Owen, R. and Macnaghten, P. (2013). 'Developing a framework for responsible innovation'. *Research Policy* 42(9):1568–1580. https://doi.org/10.1016/j.respol.2013.05.008

Stirling, A., Hayes, K. R. and Delborne, J. (2018). 'Towards inclusive social appraisal: risk, participation and democracy in governance of synthetic biology'. In: *BMC Proceedings*. BioMed Central, p. 15. https://doi.org/10.1186/s12919-018-0111-3

Stodden, V. (2009). 'The legal framework for reproducible scientific research: Licensing and copyright'. *Computing in Science & Engineering*. IEEE, 11(1). https://doi.org/10.1109/MCSE.2009.19

Stokstad, E. (2018). 'European Union expands ban of three neonicotinoid pesticides'. *Science*. https://doi.org/10.1126/science.aau0152

Stowell, S.M.L., Pinzone, C.A. and Martin, A.P. (2017). 'Overcoming barriers to active interventions for genetic diversity'. *Biodiversity and Conservation* 26(8):1753–1765. https://doi.org/10.1007/s10531-017-1330-z

Strauss, D.M. (2012). 'Liability for Genetically Modified Food'. *SciTech Law* 9:8.

Sullivan, K. (2018). *PETA Cosponsors Global Biodesign Challenge to Help "Discover" Vegan Wool*. Available at: https://www.peta.org/blog/peta-cosponsors-global-biodesign-challenge-help-discover-vegan-wool/ (Accessed: 16 July 2018).

Sunstein, C.R. (2005). 'The Precautionary Principle as a Basis for Decision Making'. *The Economists' Voice* 2(2):1–9. https://doi.org/10.1061/(ASCE)0893-1321(2003)16:1(1)

Suter II, G.W. (2016). 'Ecological Risk Assessment Frameworks'. In: *Ecological Risk Assessment*. CRC Press.

Sutton, R.I. and Staw, B.M. (1995). 'What theory is not'. *Administrative Science Quarterly*, pp. 371–384. https://doi.org/10.2307/2393788

Swainsbury, D.J.K., Martin, E.C., Vasilev, C., Parkes-Loach, P.S., Loach, P.A. and Hunter, C.N. (2017). 'Engineering of a calcium-ion binding site into the RC-LH1-PufX complex of *Rhodobacter sphaeroides* to enable ion-dependent spectral red-shifting'. *Biochimica et Biophysica Acta (BBA)-Bioenergetics* 1858(11):927–938. https://doi.org/10.1016/j.bbabio.2017.08.009

Swaisgood, R., Wang, D. and Weif, F. (2016). *IUCN Red List of Threatened Species: Ailuropoda melanoleuca*. Available at: https://doi.org/10.2305/IUCN.UK.2016-2.RLTS.T712A45033386.en (Accessed: 29 June 2018).

Synbiosafe (2018). *No Title*. Available at: http://www.synbiosafe.eu (Accessed: 16 July 2018).

Synbiowatch (2016). *A Call for Conservation with a Conscience: No Place for Gene Drives in Conservation*. Available at: http://www.etcgroup.org/files/files/final_gene_drive_letter.pdf.

Tallmon, D.A., Luikart, G. and Waples, R. S. (2004). 'The alluring simplicity and complex reality of genetic rescue'. *Trends in Ecology & Evolution* 19(9):489–496. https://doi.org/10.1016/j.tree.2004.07.003

Taylor, M.B. and Ehrenreich, I. M. (2015). 'Higher-order genetic interactions and their contribution to complex traits'. *Trends in Genetics* 31(1):34–40. https://doi.org/10.1016/j.tig.2014.09.001

Team Wageningen, iGEM (2016). *Team Wageningen*. Available at: http://2016.igem.org/Team:Wageningen_UR (Accessed: 10 July 2018).

TEDxDeExtinction (2013). *TEDxDeExtinction*. Available at: https://www.ted.com/tedx/events/7650 (Accessed: 1 July 2018).

Tershy, B.R., Shen, K-W., Newton, K.M., Holmes, N.D. and Croll, D.A. (2015). 'The importance of islands for the protection of biological

and linguistic diversity'. *Bioscience* 65(6):592–597. https://doi.org/10.1093/biosci/biv031

The Economist (2018). 'Sequencing the world'. January, pp. 70–71.

The European Group on Ethics in Science and New Technologies to the European Commission (2009). *Ethics of synthetic biology. Opinion No. 25.* Luxembourg.

The Royal Synthetic Biology Society (2017). *Synthetic Biology*. Available at: https://royalsociety.org/topics-policy/projects/synthetic-biology/ (Accessed: 16 July 2018).

The Subsidiary Body on Implementation (2018). *Global multilateral benefit-sharing mechanism (Article 10) of the Nagoya Protocol. CBD/SBI/2/5*. Montreal.

Thevenon, F., Carroll, C. and Sousa, J. (eds.) (2014). 'Plastic debris in the ocean: the characterization of marine plastics and their environmental impacts, situation analysis report'. Gland, Switzerland: IUCN. https://doi.org/10.2305/IUCN.CH.2014.03.en

Thompson, P.B. (2003). 'Value judgments and risk comparisons. The case of genetically engineered crops'. *Plant Physiology* 132(1):10–16. https://doi.org/10.1104/pp.103.022095

Thompson, P.B. (2018). 'The roles of ethics in gene drive research and governance'. *Journal of Responsible Innovation* 5(sup1):S159–S179.

Tittensor, D.P., Walpolem M., Hill, S.L., Boyce, D.G., Britten, G.L., Burgess, N.D., Butchart, S.H., Leadley, P.W., Regan, E.C., Alkemade, R., Baumung, R., Bellard, C., Bouwman, L., Bowles-Newark, N.J., Chenery, A.M., Cheung, W.W., Christensen, V., Cooper, H.D., Crowther, A.R., Dixon, M.J., Galli, A., Baveau, V, Gregory, R.D., Guitterrez, N.L., Hirsch T.L., Hoft, R., Januchowski-Hartley, S.R., Karmann, M., Krug, C.B., Leverington, F.J., Loh, J., Lojenga, R.K., Malsch, K., Marques, A., Morgan, D.H., Mumby, P.J., Newbold, T., Noonan-Mooney, K., Pagad, S.N., Parks, B.C., Pereira, H.M., Robertson, T., Rondinini, C., Santini, L., Scharlemann, J.P., Schindler, S., Sumaila, U.R., Teh, L.S, van Kolck, J. and Ye, Y. (2014). 'A mid-term analysis of progress toward international biodiversity targets'. *Science* 346(6206):241–244. https://doi.org/10.1126/science.1257484

Tompkins, D.M. (2007). 'Population bottlenecks and avian immunity: implications for conservation'. *Animal Conservation* 10(1):11–13. https://doi.org/10.1111/j.1469-1795.2006.00091.x

Tompkins, D.M., Carver, S., Jones, M.E., Krkosek, M. and Skerratt, L.F. (2015). 'Emerging infectious diseases of wildlife: a critical perspective'. *Trends in Parasitology* 31(4):149–159. https://doi.org/10.1016/j.pt.2015.01.007

Tonga National Council of Churches (2001). *Statement of Bio-Ethics Consultation*. Nuku'alofa.

Tranel, P.J. and Horvath, D.P. (2009). 'Molecular biology and genomics: new tools for weed science'. *Bioscience* 59(3):207–215. https://doi.org/10.1525/bio.2009.59.3.5

Trudeau, V.L., Schueler, F.W., Navarro-Martin, L., Hamilton, C.K., Bulaeva, E., Bennett, A., Fletcher, W. and Taylor, L. (2013). 'Efficient induction of spawning of Northern leopard frogs (*Lithobates pipiens*) during and outside the natural breeding season'. *Reproductive Biology and Endocrinology* 11(1):14. https://doi.org/10.1186/1477-7827-11-14

Tung, O.J.L. (2014). 'Transboundary movements of genetically modified organisms and the Cartagena Protocol: key issues and concerns'. *Potchefstroom Electronic Law Journal/Potchefstroomse Elektroniese Regsblad* 17(5):1739–1787. https://doi.org/10.4314/pelj.v17i5.01

Tuomisto, H.L. and Joost Teixeira de Mattos, J. (2011). 'Environmental Impacts of Cultured Meat Production'. *Environmental Science & Technology* 45:6117–6123. https://doi.org/10.1021/es200130u

Unckless, R.L., Clark, A.G. and Messer, P.W. (2016). 'Evolution of resistance against CRISPR/Cas9 gene drive'. *Genetics*, p. genetics-116. https://doi.org/10.1101/058438

United Nations (UN) (2015). *Sustainable Development Goals*. https://static1.squarespace.com/static/54a6bdb7e4b08424e69c93a1/t/597bccff4402438918153c5c/1501285648350/Bakubung-FinalReport-Web.pdf

United Nations Convention on Biological Diversity (UN CBD) (2000). *The Cartagena Protocol on Biosafety*. Available at: http://bch.cbd.int/protocol (Accessed: 16 July 2018).

United Nations Convention on Biological Diversity (UN CBD) (2015). *CBD Technical Series No. 82 Synthetic Biology*. Montreal.

United Nations Convention on Biological Diversity (UN CBD) (2017). *Portal on Synthetic Biology*.

United Nations Educational, Scientific, and Cultural Organization (UNESCO) (2015). *Ethical Perspective on Science, Technology and*

Society: A Contribution to the Post-2015 Agenda.

United States Environmental Protection Agency (US EPA) (1998). 'Guidelines for ecological risk assessment'. In: *Risk Assessment Forum*.

United States Environmental Protection Agency (US EPA) (1999). 'Phytoremediation Resource Guide'. Government of the United States of America: Washington, USA, p. 56.

United States Environmental Protection Agency (US EPA) (2017). *Final registration decision of the new active ingredient Wolbachia pipientis ZAP (wPip)strain in Aedes albopictus*. Available at: https://www.regulations.gov/document?D=EPA-HQ-OPP-2016-0205-0034 (Accessed: 16 July 2018).

United States Environmental Protection Agency (US EPA) (2017). *Update to the Coordinated Framework for the Regulation of Biotechnology*. Available at: https://www.epa.gov/sites/production/files/2017-01/documents/2017_coordinated_framework_update.pdf (Accessed: 18 July 2018).

United States Department of Agriculture (USDA) (2018a). *GAIN Report: Japan Environment Ministry Proposes Policy for Regulating Genome Editing*. Available at: https://gain.fas.usda.gov/Recent GAIN Publications/Environment Ministry Proposes Policy for Regulating Genome Editing _Tokyo_Japan_9-25-2018.pdf.

United States Department of Agriculture (USDA) (2018b). *USDA's National Institute of Food and Agriculture Invests in Research on the Implications of Gene Editing Technologies*. Available at: https://nifa.usda.gov/announcement/nifa-invests-research-implications-gene-editing-technologies (Accessed: 16 July 2018).

United States Fish and Wildlife Service (US FWS) (2014). *Endangered and threatened wildlife and plants; threatened species status for the Rufa Red Knot*. Available at: www.fws.gov/northeast/redknot/.

United States Fish and Wildlife Service (US FWS) (2017). *Endangered and Threatened Wildlife and Plants; Threatened Species Status for the Iiwi (Drepanis coccinea), Final Rule*.

Universal Bio Mining (2018). *Universal Bio Mining*. Available at: https://universalbiomining.com/.

Universidade Federal do Amazonas-Universidade do Estado Amazonas (UFAM-UEA) Brazil, iGEM T. (2016). *UFAM-UEA_Brazil*. Available at: http://2016.igem.org/Team:UFAM-UEA_Brazil (Accessed: 16 July 2018).

Vanderwerf, E.A., Burt, M.D., Rohrer, J.L. and Mosher, S.M. (2006). 'Distribution and prevalence of mosquito-borne diseases in O'ahu'Elepaio'. *Condor*, pp. 770–777. https://doi.org/10.1650/0010-5422(2006)108[770:DAPOMD]2.0.CO;2

Van Eenennaam, A L. (2017). 'Genetic modification of food animals'. *Current opinion in biotechnology* 44:27–34. https://doi.org/10.1016/j.copbio.2016.10.007

vanEngelsdorp, D., Evans, J.D., Saegerman, C., Mullin, C., Haubruge, E., Nguyen, B.K., Frazier, M, Frazier, J., Cox-Foster, D., Chen, Y., Underwood, R., Tarpy, D.R. and Pettis J.S. (2009). 'Colony Collapse Disorder: A Descriptive Study'. *PLoS one* 4(8):e6481. https://doi.org/10.1371/journal.pone.0006481

vanEngelsdorp, D., Traynor, K.S., Andree, M., Lichtenberg, E.M., Chen, Y., Saegerman, C. and Cox-Foster, D.L. (2017). 'Colony Collapse Disorder (CCD) and bee age impact honey bee pathophysiology'. *PLOS one*. Edited by G. Smagghe. Public Library of Science, 12(7):e0179535. https://doi.org/10.1371/journal.pone.0179535

Vecchiato, M., Argiriadis, E., Zambon, S., Barbante, C., Toscano, G., Gambaro, A. and Piazza, R. (2015). 'Persistent Organic Pollutants (POPs) in Antarctica: occurrence in continental and coastal surface snow'. *Microchemical Journal* 119:75–82. https://doi.org/10.1016/j.microc.2014.10.010

Veitch, C.R. and Clout, M.N. (eds.) (2002). *Turning the tide: The eradication of invasive species: Proceedings of the International Conference on Eradication of Island Invasives*. Gland, Switzerland: IUCN. https://portals.iucn.org/library/node/8175

Venter, O., Magrach, A., Outram, N., Klein, C.J., Possingham, H.P., Di Marco, M. and Watson, J.E.M. (2018). 'Bias in protected-area location and its effects on long-term aspirations of biodiversity conventions'. *Conservation Biology* 32(1):127–134. https://doi.org/10.1111/cobi.12970

Verbeke, W., Sans, P. and Van Loo, E. J. (2015). 'Challenges and prospects for consumer acceptance of cultured meat'. *Journal of Integrative Agriculture* 14(2):285–294. https://doi.org/10.1016/S2095-3119(14)60884-4

Vermeylen, S. (2010). 'Law as a narrative: legal pluralism and resisting euro-american (intellectual) property law through stories'. *The Journal of Legal Pluralism and Unofficial Law* 42(61):53–78. https://doi.org/10.1080/07329113.2010.10756642

Vestbo, S., Obst, M., Fernandez, F.J.Q., Intanai, I. and Funch, P. (2018). 'Present and Potential Future Distributions of Asian Horseshoe

Crabs Determine Areas for Conservation'. *Frontiers in Marine Science* 5(May). https://doi.org/10.3389/fmars.2018.00164

Vettori, C., Gallardo, F., Haggman, H., Kazana, V., Migliacci, F., Pilate, G. and Fladung, M. (editors) (2016). 'Biosafety of forest transgenic trees'. *Forestry* 82:209. https://doi.org/10.1007/978-94-017-7531-1

Vietnam Eliminate Dengue Project (2011). *Risk assessment of the pilot release of Aedes aegypti mosquitoes containing Wolbachia.*

Voight, M.L. and Hoogenboom, B.J. (2012). 'Publishing your work in a journal: understanding the peer review process'. *International Journal of Sports Physical Therapy* 7(5):452.

Wacker, A., Becher, P. and von Elert, E. (2002). 'Food quality effects of unsaturated fatty acids on larvae of the zebra mussel *Dreissena polymorpha*'. *Limnology and Oceanography* 47(4):1242–1248. https://doi.org/10.4319/lo.2002.47.4.1242

Wagner, N., Hochkirch, A., Martin, H., Matenaar, D., Rohde, K., Wacht, F., Wesch, C., Wirtz, S., Klein, R., Lotters, S., Proelss, A. and Veith, M. (2017). 'De-extinction, nomenclature, and the law'. *Science* 356(6342):1016–1017. https://doi.org/10.1126/science.aal4012

Wake, D.B. and Vredenburg, V. T. (2008). 'Are we in the midst of the sixth mass extinction? A view from the world of amphibians'. *Proceedings of the National Academy of Sciences* 105(Sup.1):11466-11473 https://doi.org/10.1073/pnas.0801921105

Waldram, M. S., Bond, W.J. and Stock, W.D. (2008). 'Ecological engineering by a mega-grazer: white rhino impacts on a South African savanna'. *Ecosystems* 11(1):101–112. https://doi.org/10.1007/s10021-007-9109-9

Wang, F., Wang, C., Liu, P., Lei, C., Hao, W., Gao, Y., Yao-Guang, L., Zhao, K. (2016). 'Enhanced rice blast resistance by CRISPR/Cas9-targeted mutagenesis of the ERF transcription factor gene OsERF922'. *PloS one* 11(4):e0154027. https://doi.org/10.1371/journal.pone.0154027

Wang, Y., Cheng, X., Shan, Q., Zhang, Y., Liu, J. and Qiu, J.L. (2014). 'Simultaneous editing of three homoeoalleles in hexaploid bread wheat confers heritable resistance to powdery mildew'. *Nature biotechnology* 32(9):947. https://doi.org/10.1038/nbt.2969

Warburton, B., Tompkins, D.M., Choquenot, D. and Cowan, P. (2012). 'Minimising number killed in long-term vertebrate pest management programmes, and associated economic incentives'. *Animal Welfare* 21(1), pp. 141–149. https://doi.org/10.7120/096272812X13345905674123

Wareham, C. and Nardini, C. (2015). 'Policy on Synthetic Biology: Deliberation, Probability, and the Precautionary Paradox'. *Bioethics* 29(2):118–125. https://doi.org/10.1111/bioe.12068

Warmink, J. J., Janssen, J.A.E.B., Booij, M.J., Krol, M.S. (2010). 'Identification and classification of uncertainties in the application of environmental models'. *Environmental Modelling & Software* 25(12):1518–1527. https://doi.org/10.1016/j.envsoft.2010.04.011

Webber, B.L., Raghu, S. and Edwards, O.R. (2015). 'Opinion: is CRISPR-based gene drive a biocontrol silver bullet or global conservation threat?'. *Proceedings of the National Academy of Sciences* 112(34):10565–10567. https://doi.org/10.1073/pnas.1514258112

Weinert, L.A., Araujo-Jnr, E.V., Ahmed, M.Z. and Welch, J.J. (2015). 'The incidence of bacterial endosymbionts in terrestrial arthropods'. *Proceedings of the Royal Society of London B: Biological Sciences* 282(1807):20150249. https://doi.org/10.1098/rspb.2015.0249

Weinstein, R.S. (2011). 'Should remaining stockpiles of smallpox virus (Variola) be destroyed?'. *Emerging Infectious Diseases* 17(4):681–683. https://doi.org/10.3201/eid1704.101865

Weis, V.M. (2008). 'Cellular mechanisms of Cnidarian bleaching: stress causes the collapse of symbiosis'. *Journal of Experimental Biology* 211(19):3059–3066. https://doi.org/10.1242/jeb.009597

Weiss, R., Gutmann, A. and Wagner, J. (2010). *New directions: The ethics of synthetic biology and emerging technologies*. Washington, D.C.

Welch, E., Bagley, M., Kuiken, T. and Louafi, S. (2017). *Potential implications of new synthetic biology and genomic research trajectories on the International Treaty for Plant Genetic Resources for Food and Agriculture (ITPGRFA or 'Treaty')*.

Westbrook, J. W. (2018). 'Merging backcross breeding and transgenic blight resistance to accelerate restoration of the American chestnut: The American Chestnut Foundation's breeding and selection plan 2015-2025'. Asheville, NC: The American Chestnut Foundation.

Westhoff, P. (2010). *The Economics of Food: How Feeding and Fueling the Planet Affects Food Prices*. FT Press.

Westra, J., van der Vlugt, C.J.B., Roesink, C.H., Hogervorst, P.A.M., Glandorf, D.C.M. (2016). 'Gene drives: policy report'. Rijksinstituut

voor Volksgezondheid en Milieu RIVM.

Whiteley, A.R., Fitzpatrick, S.W., Funk, W.C., Tallmon, D.A. (2015). 'Genetic rescue to the rescue'. *Trends in Ecology & Evolution*. Elsevier, 30(1), pp. 42–49. https://doi.org/10.1016/j.tree.2014.10.009

Wildlife Conservation Society (WCS) (2018). *Tiger populations recovering under effective protection in Thailand*. Available at: https://measures.wcs.org/Metric-Details/m/15.

World Health Organization (WHO) (2015). *WHO Global Technical Strategy for Malaria 2016–2030*. World Health Organisation.

World Health Organization (WHO) (2017). *World Malaria Report 2017.* World Health Organization. https://doi.org/10.1071/EC12504

World Health Organization (WHO) Global Malaria (2011). 'The use of DDT in malaria vector control WHO position statement'. *Environmental Health*, p. 9. https://doi.org/10.1093/oxfordjournals.aje.a010033

Wiener, J. (2018). 'Precautionary Principle'. In: L. Krämer and E. Orlando (eds.) *Principles of Environmental Law*, pp. 174–185. Cheltenham: Elgar Encyclopedia of Environmental Law. https://doi.org/10.4337/9781785365669.VI.13

Wiener, J.B. and Rogers, M.D. (2002). 'Comparing precaution in the United States and Europe'. *Journal of Risk Research* 5(4):317–349. https://doi.org/10.1080/13669870210153684

Wikelski, M., Foufopoulos, J., Vargas, F.H. and Snell, H.L. (2004). Galápagos birds and diseases: invasive pathogens as threats for island species. *Ecology and Society* 9(1). https://doi.org/10.5751/ES-00605-090105

Winfield, I. J. (2012). 'Review of the state of the world fishery resources: inland fisheries -Edited by R. Welcomme'. *Journal of Fish Biology* 81(6):2099. https://doi.org/10.1111/j.1095-8649.2012.03467.x

Winter, G. (2016a). 'Cultivation restrictions for genetically modified plants: On variety of risk governance in European and international trade law'. *European Journal of Risk Regulation* 7(1):120–143. https://doi.org/10.1017/S1867299X00005444

Winter, G. (2016b). 'In search for a legal framework for synthetic biology'. *Synthetic Biology Analysed*, pp. 171–211. https://doi.org/10.1007/978-3-319-25145-5_7

Winter, G. (2018). 'Substitution: From Alternatives to Ecological Proportionality'. In: L. Krämer (ed.) *Environmental Principles*. https://doi.org/10.4337/9781785365669.VI.18

Wisely, S.M., Ryder, O.A., Santymire, R.M., Engelhardt, J.E. and Novak, B.J. (2015). 'A road map for 21st century genetic restoration: gene pool enrichment of the black-footed ferret'. *Journal of Heredity* 106(5):581–592. https://doi.org/10.1093/jhered/esv041

World Intellectual Property Organization (WIPO) (2017). *Key Questions on Patent Disclosure Requirements for Genetic Resources and Traditional Knowledge*. Geneva, Switzerland: WIPO.

Woessner, J. (2004). *Faculty of 1000 evaluation for production of very long chain polyunsaturated omega-3 and omega-6 fatty acids in plants.* F1000 - Post-publication peer review of the biomedical literature.

Wolinsky, H. and Husted, K. (2015). 'Science for food: Molecular biology contributes to the production and preparation of food'. *EMBO reports*, p. e201540128. https://doi.org/10.15252/embr.201540128

Wong, M. H. (2012). *Environmental contamination: Health risks and ecological restoration*. CRC press. https://doi.org/10.1201/b12531

Woodcock, P., Cottrell, J.E., Buggs, R.J.A., Quine, C.P. (2017). 'Mitigating pest and pathogen impacts using resistant trees: a framework and overview to inform development and deployment in Europe and North America'. *Forestry: An International Journal of Forest Research* 91(1):1–16. https://doi.org/10.1093/forestry/cpx031

Woodhams, D. C., Ardipradja, K., Alford, R.A., Marantelli, G., Reinert, L.K., Rollins-Smith, L.A. (2007). 'Resistance to chytridiomycosis varies among amphibian species and is correlated with skin peptide defenses'. *Animal Conservation* 10(4):409–417. https://doi.org/10.1111/j.1469-1795.2007.00130.x

Woodrow Wilson Center (WWC) (2015). *U.S. trends in synthetic biology research funding*.

Woods, F.W. and Shanks, R.E. (1959). 'Natural replacement of chestnut by other species in the Great Smoky Mountains National Park'. *Ecology* 40(3):349–361. https://doi.org/10.2307/1929751

Woodworth, B.L., Atkinson, C.T., LaPointe, D.A., Hart, P.J., Spiegel, C.S., Twweek, E.J., Henneman, C., LeBrun, J., Denette, T., DeMots, R., Kozar, K.L., Triglia, D., Lease, D., Gregor, A., Smith, T., Duffy, D. (2005). 'Host population persistence in the face of introduced vector-borne diseases: Hawaii amakihi and avian malaria'. *Proceedings of the National Academy of Sciences*

102(5):1531–1536. https://doi.org/10.1073/pnas.0409454102

World Commission on Environment and Development (1987). *Our common future*. Oxford; New York: Oxford University Press.

World Economic Forum's System Initiative on Shaping the Future of Environment and Natural Resource Security (2018). *Harnessing the Fourth Industrial Revolution for Life on Land*. Geneva, Switzerland: WEF.

Worthy, K.A., Strohman, R.C. and Billings, P.R. (2005). 'Agricultural Biotechnology Science Compromised'. *Controversies in Science and Technology*, pp. 135–149.

Wozniak, C.A., McClung, G., Gagliardi, J., Segal, M. and Matthews, K. (2013). 'Regulation of Genetically Engineered Microorganisms Under FIFRA, FFDCA and TSCA'. In: *Regulation of Agricultural Biotechnology: The United States and Canada*, pp. 57–94. Dordrecht: Springer Netherlands. https://doi.org/10.1007/978-94-007-2156-2_4

Wright, J. (2011). 'Evaluating the use of 1080: predators, poisons and silent forests'. *New Zealand Government Report. Parliamentary Commissioner for the Environment, Wellington, New Zealand*.

Wyler, L. S. and Sheikh, P. A. (2013). 'International illegal trade in wildlife: Threats and US policy'. In: R. Gagnier (ed.) *Illicit Trade in Wildlife and the Economics of Agricultural and Wildlife Smuggling*, pp. 1–55. New York, NY: Nova Science Publishers.

Wyman, R.L. (1998). 'Experimental assessment of salamanders as predators of detrital food webs: effects on invertebrates, decomposition and the carbon cycle'. *Biodiversity & Conservation* 7(5):641–650. https://doi.org/10.1023/A:1008856402258

Wynberg, R. and Laird, S.A. (2018). 'Fast Science and Sluggish Policy: The Herculean Task of Regulating Biodiscovery'. *Trends in Biotechnology* 36(1):1–3. https://doi.org/10.1016/j.tibtech.2017.09.002

Xu, W., Vina, A., Kong, L., Pimm, S.L., Zhang, J., Yang, W., Xiao, Y. Zhang, L., Chen, X., Liu, J. and Ouyang, Z. (2017). 'Reassessing the conservation status of the giant panda using remote sensing'. *Nature ecology & evolution* 1(11):1635.

Yassif, J. (2017). *'Genspace — DIYbio Labs Project' (Open Philanthropy Project, 2017)*. Available at: https://fnih.org/what-we-do/current-lectures-awards-and-events/gene-drive-research-forum (Accessed: 25 July 2018).

Young, T. (2004). *Genetically Modified Organisms and Biosafety*. Gland, Switzerland and Cambridge, UK: IUCN. https://doi.org/10.2305/IUCN.CH.2004.PGC.1.en

Zabalou, S., Riegler, M., Theodorakopoulou, M., Stauffer, C., Savakis, C. and Bourtzis, K. (2004). 'Wolbachia-induced cytoplasmic incompatibility as a means for insect pest population control'. *Proceedings of the National Academy of Sciences* 101(42):15042–15045. https://doi.org/10.1073/pnas.0403853101

Zavaleta, E.S., Hobbs, R.J. and Mooney, H.A. (2001). 'Viewing invasive species removal in a whole-ecosystem context'. *Trends in Ecology & Evolution* 16(8):454–459. https://doi.org/10.1016/S0169-5347(01)02194-2

Zetterberg, C. and Edvardsson Björnberg, K. (2017). 'Time for a New EU Regulatory Framework for GM Crops?'. *Journal of Agricultural and Environmental Ethics* 30(3):325–347. https://doi.org/10.1007/s10806-017-9664-9

Zhang, B., Oakes, A.D., Newhouse, A.E., Baier, K.M., Maynard, C.A. and Powell, W.A. (2013). 'A threshold level of oxalate oxidase transgene expression reduces *Cryphonectria parasitica*-induced necrosis in a transgenic American chestnut (*Castanea dentata*) leaf bioassay'. *Transgenic Research* 22(5):973–982. https://doi.org/10.1007/s11248-013-9708-5

ZOA (2018). *ZOA*. Available at: http://zoa.is/ (Accessed: 11 February 2018).

Zoloth, L. (2016). 'Why wiping out female malarial mozzies is the ethical choice'. *Cosmos*, November. Available at: https://cosmosmagazine.com/society/swatting-the-malarial-mozzies.

Zügh art, W., Raps, A., Wurst-Saucy, A-G, Dolezel, M. and Eckerstorfer, M. (2011). *Monitoring of genetically modified organisms*. BfN. Federal Agency for Nature Conservation. https://www.bfn.de/fileadmin/MDB/documents/themen/monitoring/positionspapier_monitoring-gentechnik.pdf

Legal instruments and cases

African Commission on Human and Peoples' Rights (ACHPR). 2009 Centre for Minority Rights Development (Kenya) and Minority Rights Group (on behalf of Endorois Welfare Council) v. Kenya, no. 276/03.

African Commission on Human and Peoples' Rights (ACHPR). 2001. Social and Economic Rights Action Center (SERAC) and Center for Economic and Social Rights (CESR) v. Nigeria, no. 155/96.

Brazilian Legislature. 1996. Law on Industrial Property Law No. 9.279.

Council of Europe. 1993. Convention on Civil Liability for Damage Resulting from Activities Dangerous to the Environment (Lugano Convention).

Council of Europe. 1997. Convention for the Protection of Human Rights and Dignity of the Human Being with regard to the Application of Biology and Medicine: Convention on Human Rights and Biomedicine. Oviedo, 4.IV.1997. https://rm.coe.int/168007cf98

Council of Europe. 1999. Convention for the protection of Human Rights and Dignity of the Human Being with regard to the Application of Biology and Medicine: Convention on Human Rights and Biomedicine. https://www.coe.int/en/web/conventions/full-list/-/conventions/treaty/164

European Court of Justice (ECJ). Case C-528/16: Judgment of the Court (Grand Chamber) of 25 July 2018 - Confédération paysanne, Réseau Semences Paysannes, Les Amis de la Terre France, Collectif vigilance OGM et Pesticides 16, Vigilance OG2M, CSFV 49, OGM : dangers, Vigilance OGM 33, Fédération Nature et Progrès v Premier ministre, Ministre de l'agriculture, de l'agroalimentaire et de la forêt.

European Court of Justice (ECJ). Case C-165/08: Judgment of the Court (Second Chamber) of 16 July 2009 — Commission of the European Communities v Republic of Poland (Genetically modified organisms — Seed — Prohibition on placing on the market — Prohibition on inclusion in the national catalogue of varieties — Directives 2001/18/EC and 2002/53/EC — Reliance on ethical and religious grounds — Burden of proof).

European Union (EU). 1998. Directive 98/44/EC of the European Parliament and of the Council of 6 July 1998 on the legal protection of biotechnological inventions.

European Union (EU). 2000. Communication from the Commission on the Precautionary Principle. Brussels.

European Union (EU). 2001. Directive 2001/18/EC of the European Parliament & of the Council of 12 March 2001 on the deliberate release into the environment of genetically modified organisms & repealing Council Directive 90/220/EEC.

European Union (EU). 2004. EU Liability Directive — Environmental Liability with Regard to the Prevention and Remedying of Environmental Damage.

European Union (EU). 2006. Regulation (EC) No 1907/2006 — Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH).

German Federal Ministry of Justice and Consumer Protection. 2017 (amended). Genetic Engineering Act (Gentechnikgesetz - GenTG). https://www.gesetze-im-internet.de/gentg/BJNR110800990.html

Government of Canada. 2018. Food and Drug Regulations (C.R.C., c. 870). https://laws-lois.justice.gc.ca/eng/regulations/c.r.c.,_c._870/page-1.html

Government of Canada. 2018. Seeds Regulations (C.R.C., c. 1400). https://laws-lois.justice.gc.ca/eng/regulations/c.r.c.,_c._1400/index.html

Government of Japan. 2003. Act on the Conservation and Sustainable Use of Biological Diversity through Regulations on the Use of Living Modified Organisms. Act 97. https://www.env.go.jp/en/laws/nature/act_csubdrlmo.pdf

High Court of New Zealand. 2014. Sustainability Council of New Zealand Trust v. Environmental Protection Agency. Wellington Registry.

Human Rights Committee, Chief Bernard Ominayak and the Lubicon Lake Band v. Canada, Communication no. 167/1984, 38th Session, 14 February 1984, UN Doc. CCPR/C/38/D/167/1984, 1990.

Inter-American Court of Human Rights (IACHR). 2001 Case of the Mayagna (Sumo) Awas Tingni Community v. Nicaragua, Judgment of August 31, 2001 (Merits, Reparations and Costs), Series C no. 79.

Inter-American Court of Human Rights (IACHR). 2007 Case of the Saramaka People v. Suriname Judgment of November 28, 2007

(Preliminary Objections, Merits, Reparations and Costs), Series C No. 172.

Inter-American Commission on Human Rights (IACHR), Maya Indigenous Communities of the Toledo District v. Belize, Judgment of October 12, 2004 (Merits), Report no. 40/04, Case no. 12.053.

International Labour Organization (ILO). 1989. ILO Convention 169 - Indigenous and Tribal Peoples Convention. https://www.ilo.org/dyn/normlex/en/f?p=NORMLEXPUB:12100:0::NO::P12100_ILO_CODE:C169

International Court of Justice (ILC). 1995. Request for an Examination of the Situation in Accordance with Paragraph 63 of the Court's Judgment of 20 December 1974 in the *Nuclear Tests* (New Zealand v. France) Case. https://www.icj-cij.org/en/case/97

International Court of Justice (ILC). 1997. Gabčíkovo-Nagymaros Project (Hungary/Slovakia). https://www.icj-cij.org/en/case/92

International Court of Justice (ILC). 2010. Pulp Mills on the River Uruguay (Argentina v. Uruguay). https://www.icj-cij.org/en/case/135/judgments

International Law Commission (ILC). 2001. Draft Articles on Responsibility of States for Internationally Wrongful Acts. http://legal.un.org/ilc/texts/instruments/english/commentaries/9_6_2001.pdf

International Law Commission (ILC). 2006. Draft principles on the allocation of loss in the case of transboundary harm arising out of hazardous activities. http://legal.un.org/ilc/texts/instruments/english/commentaries/9_10_2006.pdf

Ministry of the Interior of North Rhine-Westphalia. 2000. Law for the Protection of Nature in North Rhine-Westphalia Land Nature Conservation Act. https://recht.nrw.de/lmi/owa/br_bes_text?anw_nr=2&gld_nr=7&ugl_nr=791&bes_id=4910&aufgehoben=N&menu=1&sg=0

People's Republic of China. 2008. Patent Law of the People's Republic of China. https://www.wipo.int/edocs/lexdocs/laws/en/cn/cn028en.pdf

Republic of South Africa. 2004. National Environmental Management: Biodiversity Act, 2004. Cape Town. https://www.environment.gov.za/sites/default/files/legislations/nema_amendment_act10.pdf

Rio Declaration on Environment and Development. UN Doc. A/CONF.151/26 (vol. I); 31 ILM 874 (1992). http://www.unesco.org/education/pdf/RIO_E.PDF

Stockholm Declaration 1972. Declaration of the United Nations Conference on the Human Environment. http://www.un-documents.net/aconf48-14r1.pdf

Supreme Court of the Philippines. 1993. Minors Oposa v. Secretary of the Department of Environmental and Natural Resources. 33 ILM 173.

Supreme Court of the United States. 2013. Assoc. for Molecular Pathology v. Myriad Genetics, Inc., 569 U.S. 576.

United Nations (UN). 1976. International Covenant on Civil and Political Rights. https://treaties.un.org/doc/publication/unts/volume%20999/volume-999-i-14668-english.pdf

United Nations (UN). 1976. International Covenant on Economic, Social and Cultural Rights. https://treaties.un.org/doc/treaties/1976/01/19760103%2009-57%20pm/ch_iv_03.pdf

United Nations (UN). 2007. Declaration on the Rights of Indigenous Peoples. https://www.un.org/development/desa/indigenouspeoples/declaration-on-the-rights-of-indigenous-peoples.html

United Nations Convention on Biological Diversity (UN CBD). 2000. Cartagena Protocol on Biosafety to the Convention on Biological Diversity. Montreal. https://www.cbd.int/doc/legal/cartagena-protocol-en.pdf

United Nations Convention on Biological Diversity (UN CBD). 2006. COP VIII Decisions. VIII/16. https://www.cbd.int/decisions/cop/?m=cop-08

United Nations Convention on Biological Diversity (UN CBD). 2012. COP XI Decisions. https://www.cbd.int/decisions/cop/?m=cop-11

United Nations Convention on Biological Diversity (UN CBD). 2016. COP XIII Decisions. XIII/17. https://www.cbd.int/decisions/cop/13

United Nations Convention on Biological Diversity (UN CBD). 2018. CBD/SBSTTA/22/4 Synthetic Biology. Montreal. https://www.cbd.int/doc/c/6e0d/b361/a877d43db3665160cce5d96e/sbstta-22-04-en.pdf

United Nations Convention on the Law of the Sea (UNCLOS). 1982. http://www.un.org/Depts/los/convention_agreements/texts/unclos/unclos_e.pdf

United Nations Economic Commission for Europe (UNECE). 1998. Convention on Access to Information, Public Participation in Decision-Making and Access to Justice in Environmental Matters (Aarhus Convention). https://www.unece.org/fileadmin/DAM/env/pp/documents/cep43e.pdf

United Nations Educational, Scientific and Cultural Organization (UNESCO). 1999. WCS Declaration on Science and the Use of Scientific Knowledge. http://www.unesco.org/science/wcs/eng/declaration_e.htm

United Nations General Assembly (UNGA). 2015. Sustainable Development Goals: Transforming Our World: the 2030 Agenda for Sustainable Development. A/Res/70/1.

United Nations General Assembly (UNGA). 2017. A/Res/72/249. http://www.un.org/en/ga/72/resolutions.shtml

United Nations (UN). 1945. United Nations Charter. http://www.un.org/en/charter-united-nations/

United Republic of Tanzania. 2009. The Environmental Management (Biosafety) Regulations. http://tz.chm-cbd.net/biosafety/national-implementation/national-biosafety-framework/tanzania-biosafety-regulations-2009.pdf

United States Code of Federal Regulations (US CFR). 2018. §340.4 Permits for the introduction of a regulated article. US 7 CFR 340.4

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