Engineered pathogens: the opportunities, risks and challenges

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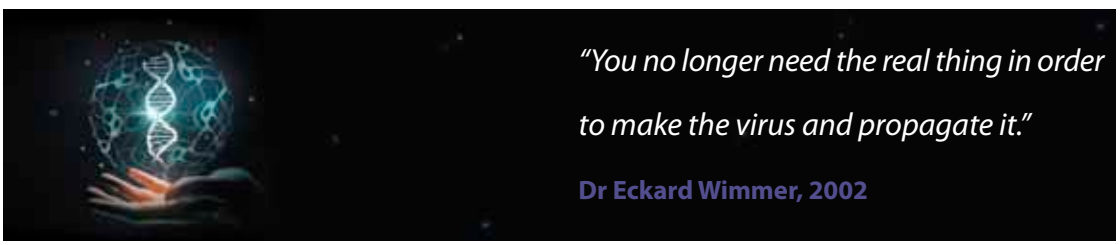
Before modern times, only nature was capable of engineering pathogens. This ability was by no means unimpressive: evolution has repeatedly demonstrated a formidable capacity for producing a vast array of infectious agents. Pathogens such as *Variola major* and *Yersinia pestis*, which cause smallpox and plague respectively, wielded enough destructive power to shape parts of human history. However, recent advancements in biotechnology mean it is now possible to engineer new viruses and bacteria. Developments in the field of synthetic biology present many exciting opportunities, enabling better understanding of disease-causing agents and facilitating the creation of new medical therapeutics and diagnostics. However, with these breakthroughs comes the risk that some of the worst pathogens in history could be recreated without requiring access to natural sources. Furthermore, engineered microbes may surpass the destructive potential of their evolved counterparts by being designed to be deadlier or more transmissible. These enhanced agents could pose an unprecedented pandemic threat to the global community. Given the risks, it is essential that regulatory frameworks for potentially hazardous research reflect modern capabilities and address emerging biosecurity concerns.

Creating synthetic life

The 21st century has seen a growing ability to apply engineering principles to biology. Viruses and bacteria can be modified with increasing ease, and some forms of life can now be created from scratch. Several milestones in microbe synthesis have been achieved relatively recently. In 2010, engineering techniques allowed an entire bacterial genome to be constructed from base material. In 2016, a synthetic living cell, named by its creators 'Syn 3.0', was created using a pared-down genome. Supported by a mere 473 genes, Syn 3.0 nonetheless has all the information necessary to support cellular life.

The ability to construct viable microbes opens the door to many opportunities. Arguably one of the greatest engineering successes to date has involved viruses that infect bacteria, known as bacteriophages. With their relatively small genome size and ease of propagation in the lab compared with eukaryotic viruses, bacteriophages have demonstrated great promise in the development of antibiotic alternatives, bacterial diagnostics and targeted drug delivery. There is a growing collection of sequenced bacteriophage genomes and catalogues of their specific gene functions. This has allowed the development of 'recombineering' (a portmanteau of **recombin**ation mediated genetic engin**eering**) techniques using bacteriophage elements to manipulate bacterial genomes in a targeted fashion.



"You no longer need the real thing in order to make the virus and propagate it."

Dr Eckard Wimmer, 2002

Biotechnology enabling synthetic biology

Recombineering techniques have proven versatile in the lab, but still possess various limitations. Recently, there has been an expanding list of new genome-editing technologies. For example, CRISPR techniques have made changing the code of life easier than ever before. This has numerous promising applications in the biomedical sciences and for research that involves manipulating pathogen genomes. The latter could help scientists better understand microbial gene functions, but also lowers the barrier to engineering 'designer bugs' with greater virulence or transmissibility than their natural counterparts.

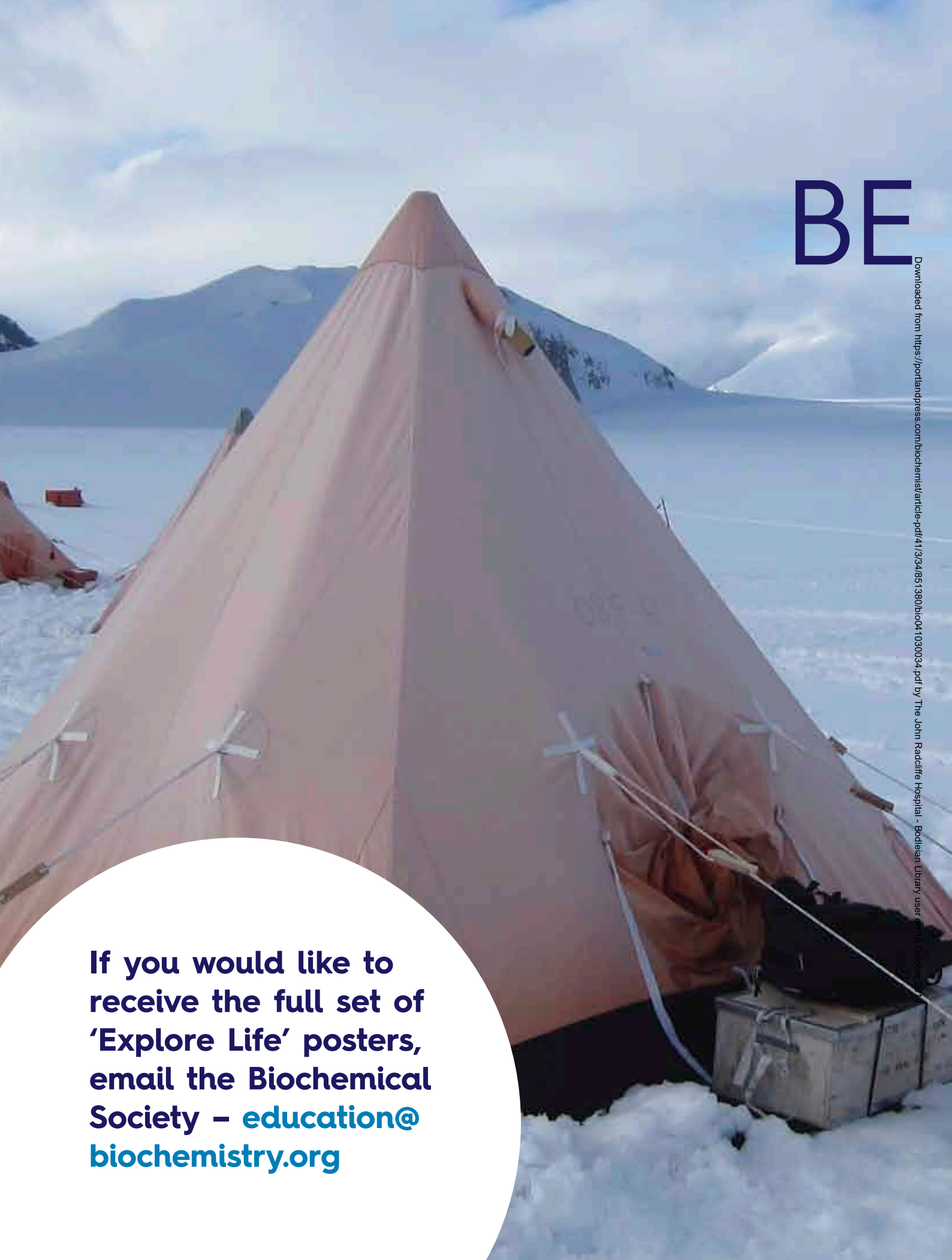
Breakthroughs in biotechnology have meant that it is now easier to synthesize genetic material faster, cheaper and more reliably than ever before. Massive parallel sequencing allows large stretches of DNA to be assembled rapidly and gene synthesis companies around the world have formed to meet the demand for orders. This has driven down cost, with the price per base pair plummeting more than 250-fold in the last 10 years alone. Lowering of the financial and technical barriers to DNA synthesis facilitates important scientific research, but also enables a broader range of actors to design and create their own pathogen genomes.

Pathogens from scratch

In 2002, a research team from the State University of New York, led by Dr Eckard Wimmer, published a paper that would revolutionize the field of synthetic biology. Using freely accessible genome data on poliovirus, Wimmer and his team placed orders from a commercial DNA synthesis company and proceeded to construct the world's first live virus from scratch. This was pioneer work in the field of synthetic biology and no mean feat at the time. Poliovirus is an RNA virus approximately 7,500 nucleotides long and a full-length complementary DNA sequence had to first be synthesized from smaller overlapping fragments before being transcribed back into viral RNA. The result was the first demonstration of a fully functional infectious virus synthesized from genomic data alone. Afterwards, in justifying the research, Wimmer stated that one of his motivations was to demonstrate that biological weapons could be created without access to natural pathogens.

Since 2002, the field of synthetic biology has expanded rapidly to include the ability to assemble more complex viral pathogens. In 2005, the pandemic 'Spanish flu' virus was reconstructed from sequenced material recovered from frozen lung tissue samples. This provided valuable insight into the biological properties of this influenza A virus which killed an estimated 50–100 million people worldwide last century. Nonetheless, this research proved controversial. In addition to the reconstruction methods, the complete genomic sequence of this deadly virus is available openly online, raising concerns that this pandemic pathogen could one day be resurrected and released, accidentally or deliberately.

More recently, research on a previously eradicated orthopoxvirus has raised new biosecurity concerns. Using mail-ordered DNA, two Canadian researchers resurrected horsepox and published their methodologies in 2018. Orthopoxviruses are some of the largest viruses known in nature, stretching more than 150,000 bp in length,



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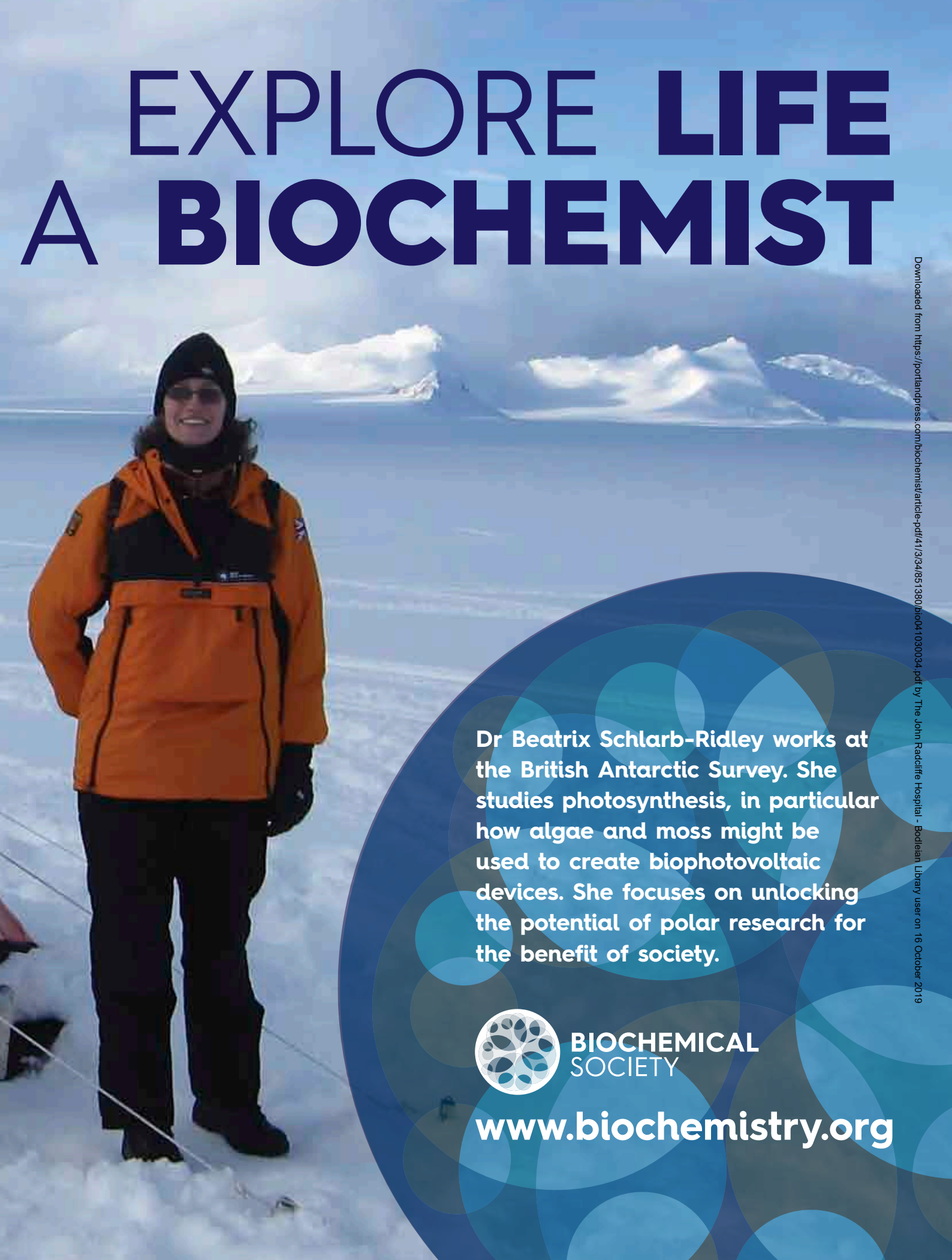

which makes their successful synthesis technically complicated. While horsepox is not a human pathogen and therefore its resurrection not a direct risk to human health, it has been argued that this research lowers the barriers for synthesizing a closely related virus, smallpox, from scratch.

Before eradication, smallpox killed 300–500 million people in the 20th century alone, more than double the number of fatalities in all wars and conflicts combined during the same time period. The world's last smallpox victim died in 1978 after contracting the virus while working on a floor above a Birmingham research facility where the virus was being handled. Nowadays, only two approved laboratories have stores of this eradicated deadly pathogen. However, with the recent publication detailing the synthesis of horsepox, there is growing concern that human pathogens like smallpox could now be created without requiring access to natural stores.

Dual-use research

Beyond pathogen synthesis, biological agents can now be enhanced in the lab through a growing list of mechanisms and techniques. It has become increasingly acknowledged that many such scientific experiments conducted on pathogens can have both good and nefarious applications. Such 'dual-use research of concern' has been defined as life sciences research that conveys benefits to society but could be reasonably anticipated to threaten public health or security if misapplied. While there are various forms of dual-use research, arguably the most controversial are gain-of function experiments which grant microbes abilities they did not previously possess.

An example of gain-of-function research, which sparked widespread discussion amongst scientists and biosecurity experts, began in 2012 when Ron Fouchier's group published a paper in *Science* detailing how they successfully modified highly pathogenic avian influenza A/H5N1 to become airborne transmissible between mammals. This type of influenza, which mainly circulates in bird populations in Southeast Asia, can cause morbidity and mortality in humans but has not previously been able to easily transmit between persons. However, Fouchier's research showed that a small handful of amino acid substitutions acquired by genetic modification in a ferret model were enough to allow this virus to become airborne transmissible.

Risks and governance challenges

Pathogens that are engineered to be more deadly or transmissible, to escape existing vaccines or to resist therapeutic treatments, could have great destructive potential if accidentally or deliberately released. Such risks provide a strong imperative for improved regulation of dual-use research. In some biotechnology industries, self-governance has already begun. For example, many gene synthesis companies now voluntarily screen DNA orders for known dangerous sequences. This largely commenced following controversy in 2006 when staff at UK newspaper *The Guardian* ordered a 78 bp length of smallpox DNA to a residential address in London. While the public was never at risk from this short viral sequence, the lack of security checks on orders spurred calls for improved governance and oversight.

"Information hazards are risks that arise from the dissemination or the potential dissemination of true information that may cause harm or enable some agent to cause harm." **Professor Nick Bostrom, 2011**

Unfortunately, it remains unclear whether current regulatory frameworks are sufficient and if emerging technologies will face even greater governance challenges. Historically, the global community has proven much better at responding reactively instead of proactively to new threats. However, to avoid unprecedented risks, effective regulation needs to be implemented before an event occurs. Oversight of dual-use research may be necessary at all stages of the research cycle, including during application, experiment and publication phases. Consensus between researchers and biosecurity experts needs to be reached if the risks are to be mitigated while safe scientific progress continues.

Hazardous information and unilateralist action

Engineering viable microbes still requires significant expertise, but the amount of tacit knowledge required to create pathogens continues to decline as services become more streamlined and information more easily accessible. This enables more actors, including some who may have malicious intent, to manufacture disease-causing agents. The growing list of dual-use publications and expanded access to online genomic and other data have resulted in a shift in the biosecurity risk landscape. It has been increasingly recognized that when it comes to creating pathogens, biological information may now pose a greater hazard to the global community than biological material itself.

Information hazards contained within scientific research are inherently difficult to assess and govern. Many pieces of biological knowledge pose direct or indirect risks, and recognized hazards need to be weighed against the numerous benefits brought by pathogen research. One difficult challenge in navigating potential

information hazards is the 'unilateralist's curse' where even if 99/100 scientists agree that a piece of research is too risky to conduct, it takes only one to disagree and publish for the information to become freely available. This asymmetry of action bias towards information dissemination poses difficult governance challenges given that it can only be addressed through universal adherence to scientific community norms for dual-use research. While there are many steps necessary to mitigate modern biological risks, the first remains wider acknowledgement and awareness of these concerns.

Conclusion

Breakthroughs in synthetic biology are occurring rapidly, with some scientists, such as Professor Dame Anne Glover, calling the 21st century 'the age of biology.' Engineered pathogens are a part of this age and will undoubtedly continue to yield many new scientific insights and beneficial biomedical applications over the coming decades. However, this progress comes with an increased ability to create disease-causing agents, some of which may be designed to be more dangerous than their natural counterparts. While some have previously said that nature is the worst bioterrorist, new technological capabilities and falling financial barriers may challenge this notion in the coming century. Synthetic biology is not required to make a biological weapon, but it does allow biological agents to be developed and enhanced without access to natural pathogens.

Given the complex challenges in regulating dual-use research, and the importance of promoting safe and responsible scientific community norms, more dialogue between scientists and biosecurity experts should be championed. Misuse of research and published information is a credible risk that requires a coordinated approach that avoids unilateralist actions. Taking steps that begin with improving awareness of dual-use concerns, scientists and biosecurity experts can work together to ensure that the benefits synthetic biology promises can be delivered without evoking the risks. ■

Further reading

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