

Chapter 6

DNA SYNTHESIS AND SECURITY

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1. INTRODUCTION

Progress in life sciences has given rise to fears that the related technologies could be used for malicious purposes. In particular the novel approaches and technologies that evolve from synthetic biology have alarmed policymakers and the security community and stimulated scientific, regulatory and public debate. Blurring the lines between chemistry, biology and engineering, synthetic biology enables an ever more intentional design of genetic information, biological parts and systems. This is highly relevant for biosecurity because the very idea of biological weapons roots in the possibility to control the impact and functioning of harmful biological systems. Synthetic biology clearly has the potential to help design and build so far unknown biological systems and molecular structures with very specific characteristics and impacts. As a second concern, one that seems more feasible based on the current state of the art, this field may also dramatically increase the accessibility of select agents and pathogenic organisms, thereby intensifying the proliferation threat. Especially DNA synthesis technologies reduce the hurdles to obtaining pathogens by transforming the issue successively from a question of physical access to a mere question of access to the sequence information.

This chapter will briefly describe the extent to which these threats are real; i.e. we provide a brief overview on relevant achievements in gene synthesis and relate them to other less-developed fields of synthetic biology. The main focus will, however, be on describing the current state of relevant regulations. This will include some recent developments and progress as well as concluding remarks.

2. SECURITY ISSUES RELATED TO DNA SYNTHESIS

2.1. DNA Synthesis as Part of Synthetic Biology

Although the term “synthetic biology” was already used about 100 years ago by Stephane Leduc in 1912 (Campos 2009), the contemporary version is a relatively young field at the intersection of biology, engineering, chemistry and information technology. Typical for an emerging science and engineering field, a variety of definitions are circulating in the scientific community, and no one definition would receive total support by the researchers involved in SB activities (Schmidt and Pei 2010). The probably least contested definition can be found on the SB community webpage <http://syntheticbiology.org/>.

Accordingly, “Synthetic Biology is: A) the design and construction of new biological parts, devices, and systems; and B) the re-design of existing, natural biological systems for useful purposes.

Synthetic biologists are currently working to

- specify and populate a set of standard parts that have well-defined performance characteristics and can be used (and re-used) to build biological systems,
- develop and incorporate design methods and tools into an integrated engineering environment,
- reverse engineer and re-design pre-existing biological parts and devices in order to expand the set of functions that we can access and program,
- reverse engineer and re-design a 'simple' natural bacterium,
- minimize the genome of natural bacteria and build so-called protocells in the lab, to define the minimal requirements of living entities, and
- construct orthogonal biological systems, such as a genetic code with an enlarged alphabet of base pairs.”

The lack of a well-accepted definition, however, does not prevent the community from forging ahead and doing SB, naturally leading to a quite diverse area of science and engineering. Activities that fall under SB are currently performed in several fields. For various reasons, these activities are not always addressed under the term proper, while others use the label for yet different endeavours. By and large, however, the following activities are usually subsumed under SB (Schmidt and Pei 2010)

- DNA synthesis (or synthetic genomics)
- Engineering DNA-based biological circuits (based on metabolic engineering but using real engineering principles)
- Defining the minimal genome (or minimal cell)
- Building protocells (or synthetic cells)
- Xenobiology (aka chemical synthetic biology)

2.2. Malicious Use by Non-State Actors and Terrorists

According to the WHO (2004), biosafety is the prevention of *unintentional* exposure to pathogens and toxins, or their accidental release, whereas biosecurity is the prevention of loss, theft, misuse, diversion or *intentional* release of pathogens and toxins. The consolidation of the research field synthetic biology came immediately after September 11, 2001, and the US Anthrax letters. With former US secretary of defense Colin Powell's presentation before the UN security council on Irak's alleged weapons of mass destruction program, especially the account of the (fictitious) mobile bio-weapon units, the scene was set for a rigid scrutiny of biotech R&D.

Increasing concerns in the US that research in the life sciences might be misused for bio-terrorist or bio-warfare purposes were fuelled by a number of experiments that triggered substantial debate. In particular, three experiments prompted such debates (Kelle 2007):

- Non-intentionally enhancing the virulence of the mousepox virus by inserting an IL-4 gene into the mousepox genome (Jackson et al. 2001)¹. While this experiment unexpectedly yielded a killer mousepox virus, subsequent work by another scientist, Mark Buller at Saint Louis University, has knowingly carried these experiments one step further by increasing the lethality of the mouse pox virus and by carrying out similar manipulations in the cowpox virus (Buller 2003).²
- Synthesis of the poliovirus genome from 'chemically synthesized oligo-nucleotides that were linked together and then transfected into cells, thereby creating an infectious virus from scratch, combining knowledge of the viral DNA with assembly of the correct chemical compounds (Cello et al. 2002).³
- The 1918 Spanish Flu was reconstructed in 2005.
- Transfer of the virulence factor of variola major (which causes smallpox) into the vaccinia virus, which is of much lower virulence and usually used for vaccinations against smallpox (Rosengarden et al. 2005).⁴

These experiments drew the attention of the security community to synthetic genomics and synthetic biology. Since then US security institutions and think tanks, among them the NSABB – the National Security Advisory Board on Biotechnology (http://oba.od.nih.gov/biosecurity/about_nsabb.html), the Strategic Assessment Group of the National Academy of Sciences (A darker Bioweapons Future 2003), the FBI (e.g. arresting innocent biotech-artist Steve Kurtz for having biotech equipment in his house, pre-emptive

¹ R. J. Jackson et.al. 2001, 'Expression of mouse interleukin-4 by a recombinant ectromelia virus suppresses cytolytic lymphocyte responses and overcomes genetic resistance to mousepox', *Journal of Virology*, 75 pp.1205-1210; R. Nowak 2001, 'Disaster in the making. An engineered mouse virus leaves us one step away from the ultimate bioweapon', *New Scientist* (13 January 2001) pp.4-5.

² M. Buller 2003, The potential use of genetic engineering to enhance orthopoxviruses as bioweapons. Presentation at the International Conference 'Smallpox Biosecurity. Preventing the Unthinkable' (21-22 October 2003) Geneva, Switzerland; J.D. Steinbruner, E.D. Harris 2003, 'When science breeds nightmares', *International Herald Tribune* (3 December 2003); D. MacKenzie, 'US develops lethal new viruses', *New Scientist*, 180

³ J. Cello, A.V. Paul, E. Wimmer 2002, 'Chemical synthesis of poliovirus cDNA: generation of infectious virus in the absence of natural template', *Science*, 297 pp.1016-1018.

⁴ A. M. Rosengard et.al. 2002. 'Variola virus immune evasion design: expression of a highly efficient inhibitor of human complement', *Proceedings of the National Academy of Sciences USA*, 99 pp.8808-8813.

investigation of, and attempts to cooperate with, the SB Do-It-Yourself (DIYBio) community), the Commission on the Prevention of Weapons of Mass Destruction, Proliferation and Terrorism (Graham et al. 2008⁵), the National Academies, and others.

3. CURRENT STATE OF THE REGULATORY ENVIRONMENT

On the international level, the most important regulatory regime for biosecurity is the 1972 Biological and Toxins Weapons Convention (BWC). Although it does not specifically target potential threats posed by synthetic biology, the Convention and related national and international export control regimes are setting the playing field for all synthetic biology innovation. Potentially relevant on the international level are moreover the Chemical Weapons Convention (CWC) and the Convention on the Prohibition of Military or Any Other Hostile Use of Environmental Modification Techniques (ENMOD) as well as the 2006 WHO Laboratory Biosecurity Guidance.

The starting point of international biosecurity efforts has been the 1925 Geneva Protocol, which first prohibited the deployment of chemical and biological weapons following the horrible impact of chemical warfare in World War I. With the rapid scientific progress, however, biological agents became a growing concern in the third quarter of the twentieth century, leading to the 1972 international Biological and Toxins Weapons Convention (BWC), including prohibitions on development, production and stockpiling of related materials and substances. After entering into force in 1975 the Convention was successively joined by the majority of States, counting currently more than 160 State parties.

As a strong international norm, which was never publicly challenged, the BWC “[...] unequivocally covers all microbial or other biological agents or toxins, naturally or artificially created or altered, as well as their components, whatever their origin or method of production [...]” (BWC, Additional Understanding of Article 1). Going beyond misuse of biological material occurring in nature, the Convention is also coping with artificial forms and hence already covers virtually all developments in the fields of genetic modification and DNA synthesis (Kelle 2007a, 5). Besides this comprehensive future-proof definition, another asset of the Convention is the clear ban of any but peaceful use of biology. Nonetheless, with the growing need for defensive research and preparedness for outbreaks of diseases, some of the weaknesses of the Convention’s exemptions cast doubt to the effectiveness of its implementation. In particular, the Convention lacks an organisation or implementing body or any other effective means of systematically monitoring implementation or compliance. A mechanism for investigating alleged violations is also missing. Moreover, no systematic assessment of needs or provision of assistance has been established, resulting in fairly uneven national implementation (Lennan 2010). Finally the Convention’s focus on state-based BW programs does not adequately reflect the growing role of private (non-state) actors in relevant research activities or the potential threat of bioterrorism.

Earlier initiatives, especially in the field of verification, have not gained the necessary support for substantive progress, and dissent at the 5th BWC Review Conferences (RC) in 2001 resulted in a near deadlock. The review process of the Convention nonetheless saw some revival at the 6th RC in 2006, which took account of the emerging and converging

⁵ Graham et al. 2008. *World at Risk*. Random House, New York

technology fields. In its ‘Contributions to the Science and Technology Background’, the Netherlands highlighted potential shortcomings of the Convention resulting from the extension of engineering capabilities related to bioactive nanostructures (nanotechnology). It warned that the “degree of artificiality might exclude the technology from the Convention”, recommending to include a provision in the Additional Understandings “to the effect that misuse of scientific and technological developments in the field of nanotechnology and derived applications is in fact a violation of Article I”(BWC/CONF.VI/ NL S&T; 9).

It has not been a problem to extend the Additional Understanding of the definition used in the Convention to cope with novel technological developments. Other issues embody a greater dilemma. The United States pointed to the problem of open access to data on combinatorial chemistry. Those libraries “could also be quickly and easily searched by those with malign intent for compounds with the potential to interact with endogenous physiological pathways for use as biochemical weapons.”(BWC/CONF.VI USA S&T, 2006). Given the technological progress in the field of DNA synthesis, this concern extends to data on genetic information. Note, however, that any restrictions in accessing information would inevitably interfere with peaceful research and scientific freedom and research progress.

Although the focus in nonproliferation still lies on measures to prevent unimpeded access to hazardous materials, synthetic biology is shifting the challenge to access of information, material and equipment. This approach transforms technology-based threats to knowledge-based risks. This adds to the concern already put forward by Germany at the 6th RC (BWC/Conf.VI/WP.2, 2006). Article IV emphasizes the State parties’ obligations to undertake all necessary measures to prevent any of the prohibited activities within their territories, but gives no explanations as to what these measures actually would include. This vagueness is particularly problematic in the field of synthetic biology because its scientific progress, as an enabling technology (such as DNA synthesis), is now largely driven by private, research-oriented enterprises. Beyond the reluctance of these companies to undergo additional control, most states might simply lack the expertise for effective oversight of all the rapid progress and diversification in life sciences.

For the 7th RC in 2011, it will be interesting to see whether the growing challenges from emerging technologies finally translate into a more institutionalised approach to biological weapons and biosecurity, going beyond the Implementation Support Unit (ISU) that has worked towards consensus during the past years.

Another decisive topic on which the international community will need to find common ground is the further development of confidence-building measures among member states, including their capabilities for defence and in emerging fields such as synthetic biology.

Potential conflicts gravitate on (1) technology transfer for peaceful uses (Article X of the Convention) and (2) the question of compliance. The latter issue has already proven its divisive potential between those states that consider some sort of verification necessary and others that doubt the effectiveness of any such system.

The Convention builds on a strong normative consensus with appropriately encompassing definitions that cannot easily be circumvented by synthetic biology innovations. Nonetheless, the means of implementation and verification still need to be developed to catch up with technological progress. Moreover, efforts to convince other States to sign and ratify the Convention are important to ensure its long-term success. This is even more important concerning the above-mentioned ENMOD Convention, which so far counts only 75 State parties. ENMOD could potentially become relevant to prevent deployment of harmful genes,

biological parts, devices and systems (selectively) altering and damaging the environmental, agricultural and economic base. It could prevent hostile abuses of synthetic biology innovations even if they might not classify as biological weapon.

In addition to the conventions, the United Nations Security Council in 2004 unanimously adopted Resolution 1540 under Chapter VII of the United Nations Charter. It obliges States to refrain from supporting by any means non-State actors from developing, acquiring, manufacturing, possessing, transporting, transferring or using nuclear, chemical or biological weapons and related delivery systems. The Resolution imposed binding obligations on all States and established a Committee to report on the progress and achievements. Security Council Resolutions 1673 (2006) and Resolution 1840 (2008) extended the Resolution and the Committee by two and three years, respectively. While the reports revealed some progress, the Committee also calls for continued long-term efforts. A comprehensive Committee report on achievements and shortcomings is expected for April 2011.

3.1. Australia Group and Export Control Regimes

As a reaction to the deployment of chemical agents by the Iraqi government in the 1980s, an Australian-led initiative towards informal cooperation on the identification and control of dual-use exports resulted in the establishment of the Australia Group in 1985. The group, which from the beginning encompassed all major western countries, now counts 41 states including all EU members. Its major purpose is to work for non-proliferation of chemical and biological weapons and related capabilities.

The Australia Group provides its members with control lists and guidelines supporting harmonized standards for exports. The lists encompasses a wide range of equipment related to biological weapons capabilities. It includes complete containment facilities at Biosafety Level (BSL) 3 or 4 containment level, fermenters, centrifugal separators, cross (tangential) flow filtration equipment, freeze-drying equipment, protective and containment equipment, aerosol inhalation chambers and spraying or fogging systems as well as components thereof. No specific reference has so far been made to gene synthesis machines (oligonucleotide synthesizers). A list containing select agents is kept updated and now includes genetic elements and genetically-modified organisms containing any nucleic acid sequences associated with pathogenicity of any of the microorganisms in the list or coding for any of the toxins in the list. Regular reviews are conducted by a technical advisory group that recently proposed some additions relating to emerging technologies (Australia Group, 2010).

Together with the guidelines for export controls, the provisions of the Australia Group make some exemptions for basic research. This allows some degree of reconciliation between its nonproliferation objective and Article X of the BWC, which calls for technology transfer for peaceful purposes among member states. The problem with Article X of the BWC, however, resides in the very nature of dual-use technologies.

At the European level the guidelines provided by the Australia Group are implemented by Council Regulation EC No.1334/2000, which establishes a European regime to control transfer, brokering and transit of dual-use technologies and items. Amendments and updates of this binding regime were subsequent Council Regulations EC No 2889/2000, EC No 458/2001, EC No. 2432/2001, EC No 880/2002, EC No. 149/2003, EC No. 1504/2004, EC No. 394/2006, EC No. 1183/2007 and EC No.1167/2008. In its current version (EC No.

428/2009) the Council Regulation includes in Annex I a list of human, animal and plant pathogens and toxins similar to the select agents list of the Australia Group. No reference is made to oligonucleotide synthesizers and other equipment specific to synthetic biology. Annex II of the Council Regulation specifies an authorisation of dual-use exports to Australia, Canada, Japan, New Zealand, Norway, Switzerland and the United States of America – all members of the Australia Group. Among other items, pathogens and toxins are exempted from this authorisation, and export controls apply.

The European Commission has so far not announced any specific framework dealing with security concerns related to synthetic biology. Nonetheless, a set of consultations and workshops has been held and it seems likely that the latest results of the biosecurity efforts in the US and by the industry will finally have some “spill over effect” on the EU.

In the United States, toxins and microbial organisms that have the potential to pose a severe threat to the public are regulated through the Select Agent Regulation (SAR), administered by the Department of Health and Human Services/Centers for Disease Control and Prevention (HHS/CDC) and the Department of Agriculture/Animal and Plant Health Inspection Service (USDA/APHIS). The select agents referred to in the SAR correspond to the lists provided by the Australia Group. Under the Export Administration Regulations (EAR), the Australia Group Lists are taken up in the Commerce Control List (CCL). Similar to the European regulation, the EAR includes some flexibility on dual-use exports depending on destination quantities and purpose. Additionally, the Arms Export Control Act (22 U.S.C. 2778) and the related International Traffics in Arms Regulation (ITAR) impose obligations on sensitive items to require export licenses on a case by case basis.

Contrary to the situation in the EU, security issues posed by synthetic biology attracted early and continued attention. George Church, a leading researcher in the field of synthetic biology, highlighted major concerns in “A Synthetic Biohazard Non-proliferation Proposal” as early as 2004 (Church 2004). Most of his propositions concern the synthesis of oligonucleotides as a centerpiece of artificial DNA production; they proved particularly relevant for policy approaches:

- Sequence screening for select agents to avoid synthesis of known pathogens or toxin-related DNA;
- customer screening to avoid shipment to dubious clients and
- licensing of equipment and substances required for the synthesis of oligonucleotides.

These issues have structured both the debate and its practical outcomes, and have entered all milestone reports, including the work of

- the Fink Committee of the US National Academies of Sciences on Research Standards and Practices to Prevent the Destructive Application of Biotechnology, chaired by Gerald R. Fink;
- the Lemon-Relman Committee (Report by the Committee on Advances in Technology and the Prevention of Their Application to Next Generation Biowarfare Threats, 2006);
- the Declaration of the Second International Meeting on Synthetic Biology (SB 2.0) on Biosecurity;

- the CSIS-MIT-Venter Report on Synthetic Genomics: Options for Governance (Garfinkel et al. 2007); and
- the Controlling Dangerous Pathogens project (Steinbruner et al. 2007)

More recently, the Department of Health and Human Services has proposed and established a “Screening Framework Guidance for Synthetic Double-Stranded DNA Providers“ in order “to reduce the risk that individuals with ill intent may exploit the commercial application of nucleic acid synthesis technology to access genetic material derived from or encoding Select Agents or Toxins” (Fed. Regist. 74, 62319–62327). In an attempt to reconcile the security concerns with commercial interests of gene synthesis companies, consultations with concerned stakeholders were held in early 2010.

The final version of the non-binding Framework Guidance was published in October 2010. It proposed a screening framework for commercial providers of synthetic double-stranded DNA with a length of more than 200 base pairs, responding to concerns associated with the potential for misuse of their products. Its principles and ideas compare to industry self-regulation initiatives discussed below, although its provisions are somewhat less tight.

The process leading the Framework Guidance was largely stimulated by the National Science Advisory Board for Biosecurity (NSABB). After a widely recognized report on synthetic genomics in 2006, the NSABB submitted a “Draft Report Addressing Biosecurity Concerns Related to Synthetic Biology” in April 2010. This Report calls for establishing an oversight framework for dual-use research and provides an “assessment of any biosecurity concerns presented by the ability to synthesize new genes, biochemical pathways, and biological components with specified or novel properties”. More specifically it advocates to

- make synthetic biology subject to institutional review and oversight because it identifies some aspects of this field as posing biosecurity risks;
- extend oversight of dual-use research beyond the boundaries of life sciences and academia, taking account of the growing number of synthetic biology practitioners without academic background or affiliation;
- develop education strategies addressing dual-use research issues in order to increase sensitivity to biosecurity concerns in research communities related to synthetic biology and;
- monitor advances in synthetic biology on a regular basis.

3.2. Industry Self-Regulation

When companies first started to commercially produce and sell synthetic DNA in 1999, a cycle of increased investments in R&D and equipment was initiated. This made artificial DNA sequences much more affordable than before and increased demand (Carlson 2009). At the same time, falling prices also diminished returns (Maurer et al. 2009, 1). While the market for gene synthesis will certainly see further growth in coming years, the prediction is that consolidation on the one hand and innovations on the other will continuously change the market, its players and their positions.

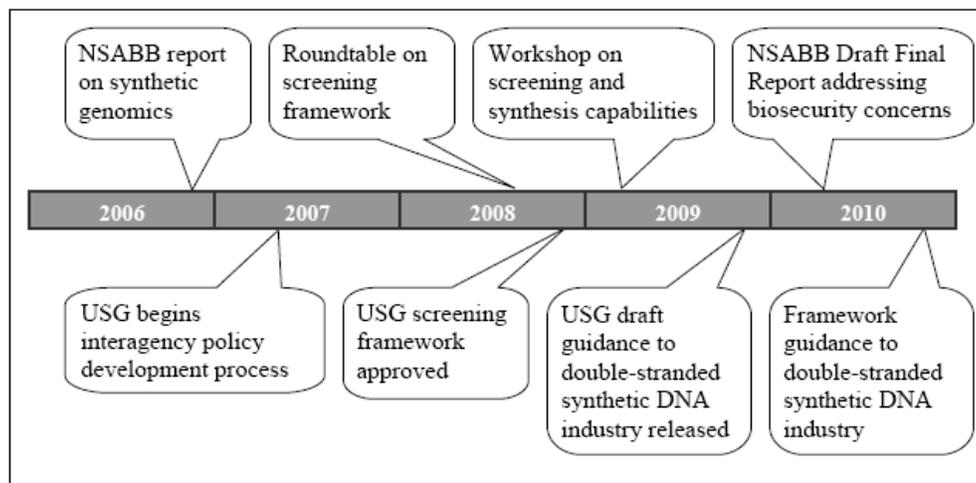


Figure 1. Recent timeline for US reports and achievements on security of synthetic biology.

Although they have acknowledged potential biosecurity issues, most companies were initially reluctant to accept the burden of additional security regulations. This position has changed successively in recent years, resulting in two comparable sets of self regulations by competing industry associations.

Both regulations set standards on sequence screening that are higher than those of the “Screening Framework Guidance for Synthetic Double-Stranded DNA Providers” published by the US Department of Health and Human Services/NIH. While this seems to astonishing at first glance, there are good reasons for the industry to endorse tougher sequence and customer screening, including:

- diminishing costs for screening through automated screening and use of algorithms,
- the risk of backlashes through proven abuses of synthesis capabilities, and
- strategic influence on any further state-driven regulation through control of the standardization process.

The idea of industry self-regulation was picked up by the International Association Synthetic Biology (IASB) and the International Gene Synthesis Consortium (IGSC). Unlike the IGSC, which restricts membership to companies with more significant market shares, the IASB, mainly driven by smaller German gene synthesis providers, is open to all companies.

Shortly after the IASB announced its intention to draft a “Code of Conduct for Best Practices in Gene Synthesis” (IASB 2009), some companies proposed a competing and less costly approach. DNA 2.0 and Geneart, both members of the IGSC, proposed significantly lower requirements for sequence screening, putting emphasis on fast and cheap computerized checks against a predefined list of threats. It soon became clear, however, that the IGSC would follow the more robust approach taken by IASB, including extended screening if necessary.

Table 1. Comparison of the three major competing DNA synthesis standards

IASB Code of Conduct for Gene Synthesis	IGSC Harmonized Screening Protocol	U.S. DHHS - NIH Screening Framework Guidance
DNA sequences submitted as inquiries or orders for DNA synthesis will be screened against Genbank for reasonable sequence similarity to pathogens. A joint Technical Experts Group on Biosecurity (TEGB) may take further reasonable steps of inquiry if considered necessary.	IGSC companies use automated screening as a filter to identify pathogen and toxin DNA sequences. When a potential pathogen or toxin sequence is identified, the order is reviewed by an expert and is either accepted, with a requirement for additional customer review, or rejected.	The purpose of sequence screening should be to identify whether “sequences of concern” are ordered, in which case further follow-up procedures should be used to determine if the order would raise concern. Automated screening is recommended for all double-stranded DNA orders.
Customer screening effort will depend on the sequence screening results. Reasonable efforts to determine the legitimacy of a customer (compliance with trade restrictions) and delivery address will be made, and records on his/her contact details kept.	Potential customers are screened against available lists provided by state authorities to check whether any concerns regarding a customer, especially if the customer is an individual, exists.	Providers should develop customer screening mechanisms to verify the legitimacy of a customer. If the customer is an individual, providers should identify potential ‘red flags’, and to conform to U.S. trade restrictions and export control regulations.
Documentation shall include records for eight years on suspicious inquiries and positive screening hits. Moreover, statistical records on the total number of inquiries and orders for synthetic genes, the number of inquiries and orders with positive screening hits, and related decisions.	Retaining records for eight years of (1) sequence screen results, (2) customer screen results and (3) product and delivery information, including at least (a) the synthetic DNA sequence; (b) the vector; and (c) the recipient’s identity and shipping address.	The Guidance recommends retaining records of (1) sequence screen results, (2) customer screen results and (3) eventual follow-up screening. Records should be kept for eight years.
Gene synthesis providers shall promptly inform authorities each time they encounter evidence which clearly suggests possible illegal activities. Such evidence will include, by way of example, inquiries and orders that strongly suggest illegal activities, such as attempts to conceal a non-business delivery address.”	IGSC companies will report any request for a gene associated with pathogens and received from a suspicious potential customer failing to establish its bone fides.	Follow-up screening is to verify the legitimacy of customers both at the level of the customer and the principal user, to confirm that customers and principal users placing an order are acting within their authority, and to verify the legitimacy of the end-use.
The Code of Conduct is binding to its IASB-signatories but also meant as a guideline for non IASB companies.	The Harmonized Screening Protocol is considered as a binding standard for IGSC members.	The Screening Framework Guidance is a nonbinding best practice guideline.

Although the three approaches to screening seem quite similar, they differ in detail. Especially the 'Best Match' method recommended by the Guidance Framework would require

performing a follow-up investigation only if a sequence were found to be more closely related to a select agent than to any other known sequence that is not considered to be a select agent (Fischer et al. 2010, 21). Moreover, the Guidance Framework and IGSC put somewhat more emphasis on customer screening.

One problem with all three standards is their somewhat limited outreach. It might well be that most of the industry will soon converge its practices to one of these model standards, and that large customers will even endorse further standardization. This will not, however, solve the problem of ever cheaper and more readily available sources for synthetic DNA. Another problem currently untouched by the regulations is individual clearance, licensing of tools and access control.

3.3. Comprehensive Approaches and Responsibilities of Scientists

The emerging industry standards and the NSABB Proposed Framework for the Oversight of Dual Use Life Sciences Research should be seen as important building blocks for security governance of synthetic biology. They are, moreover, useful means to raise awareness about security concerns in the (academic) synthetic biology community. In an attempt to develop a more comprehensive overarching governance strategy, Kelle (2009) suggested to look at the challenge from the perspective of points for potential policy impact. In his 5P-Strategy, he identified five different policy intervention points, namely the:

- principal investigator (PI), the
- project, the
- premises, the
- provider (of genetic material) and, its
- purchaser

Kelle argues that focussing on policy intervention points would enrich the ‘options for governance’ proposed by the report of the Massachusetts Institute of Technology (MIT), the Center for Strategic and International Studies (CSIS) and the J Craig Venter Institute (JCVI) (Garfinkel et al. 2007).

In particular the focus on principle investigator, project and premises could help avoid both too narrow and too broad a picture of what synthetic biology is actually about (Kelle 2009; 88).

Finally, the 5P-Strategy avoids reducing the security concerns of synthetic biology to commercial capabilities that the provider-purchaser perspective tends to imply. Instead, it allows putting more emphasis on the role of the researcher and his/her research environment and agenda.

While basic research is protected by the freedom of sciences, researchers also have legislated responsibilities and must follow restrictions, notably to avoid any violation to the provisions of the BWC and other international and national applicable law.

4. EMERGING CHALLENGES

In the next few years a number of new challenges to biosecurity and DNA-synthesis are likely to appear. One that has been discussed deals with so-called “split orders”. Split orders are the alleged action of a malintent person or organization that tries to circumvent detection systems of DNA synthesis companies by splitting up one piece of harmful DNA into many smaller, harmless-looking pieces and ordering them from a variety of different companies. Today, companies do not necessarily share the details of orders they receive, creating a clear opportunity for a successful split up. Obstacles towards integration of orders among all relevant DNA synthesis companies are mainly rooted in customer privacy. What is missing now is a trustworthy institution or system that would handle information from all DNA synthesis companies to detect split orders, without counteracting privacy concerns.

Another upcoming issue involves outsourcing. The first step of outsourcing is the actual order for custom-made DNA made to DNA synthesis companies (e.g. in the US or Europe). The resulting bottleneck favors (self-)regulation of these companies as an effective tool to prevent biosecurity threats. The second step is related to the fact that DNA synthesis companies are confronted with falling prices and costs for synthesis, along with decreasing profit margins; this leads to a second wave of outsourcing to highly specialized and highly price-competitive DNA synthesis facilities, e.g. in China or other BRIC⁶ countries. Operation of these very-high-throughput facilities in non-Australia Group countries could pose a challenge to the current regulatory system. The degree will depend very much on how these facilities and the countries in which they are located (co-)operate and react to potential biosecurity threats.

The final issue that may raise security issues in the mid- to long-term future is the potential for non-natural biological systems. Herdewijn and Marliere (2009) and Schmidt (2010), for example, describe the increasing interest in nucleic acids that do not occur in nature but have similar properties as DNA or RNA. The so-called xeno-nucleic acids (XNA) contain a different structural molecule in their chemical backbone. This yields information-storing biopolymers such as HNA (Hexose nucleic acid), TNA (Threose nucleic acid) or GNA (Glycol nucleic acid). XNAs do not fall under any DNA synthesis regulation, which is not yet a problem because XNAs are currently not produced by DNA synthesis company. Once XNAs become a commodity, similar to DNA is today, then of course XNA should be included in the DNA synthesis regulations. In contrast to the physical exchange of a molecule, the so-called code engineering deals with a change in the genetic code itself (Budisa, 2004; Cropp et al., 2007; Wang et al., 2006; Wang et al., 2009). A protein can be predicted from the sequence of base pairs on the DNA. Current software programs used by DNA synthesis companies compare the ordered DNA from customers to a database containing select agents (see text above). If a customer were able to change the universal genetic code (i.e. the translation of base pairs to amino acids and proteins) in a production host, then a DNA for a toxic protein could be ordered without the software being able to detect a security problem.

⁶ BRIC: Brasil, Russia, India, China

CONCLUDING REMARKS

Since the onset of the DNA synthesis market, specialized companies were created to satisfy the requirements of that market. These companies provided a convenient point of action for security regulations and self-imposed code of conducts to impede the outsourced production of harmful biological weapons and toxins. Despite the co-existence of several guidelines for DNA synthesis (companies), the overall field can be regarded as being under good control from a security point of view. Certain open question remain, however, and a pragmatic approach that seeks to tackle legislative and regulatory shortcomings at all levels and at any possible point of intervention is advisable. This would be a promising strategy to cope with any eventuality (such as split orders, second-level outsourcing or the onset of novel biological systems) that future innovations might unveil.

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REFERENCES

- Australia Group webpage (2010) List of biological agents for export control, November 2010. http://www.australiagroup.net/en/biological_agents.html.
- Australia Group webpage (2009) Control List of Dual-use Biological Equipment and Related Technology and Software, September 2009. http://www.australiagroup.net/en/dual_biological.html.
- Budisa, N. (2004). Prolegomena to future experimental efforts on genetic code engineering by expanding its amino acid repertoire. *Angew Chem Int Ed Engl* 43, 6426-63.
- Buller, M. (2003), The potential use of genetic engineering to enhance orthopoxviruses as bioweapons. Presentation at the International Conference ‘Smallpox Biosecurity. Preventing the Unthinkable’ (21-22 October 2003) Geneva, Switzerland.
- Campos L (2009) That was the Synthetic Biology that was. Chapter 1 in: Schmidt M. Kelle A. Ganguli A, de Vriend H. (Eds.) 2009. *Synthetic Biology. The Technoscience and its Societal Consequences*. Springer Academic Publishing.
- Carlson, R (2009). The changing economics of DNA synthesis. *Nat Biotechnol.* 27(12):1091-4.
- Cello, J. Paul JW. Wimmer E. (2002) Chemical synthesis of poliovirus cDNA: generation of infectious virus in the absence of natural template. *Science*, 297 pp.1016-1018.
- Church, G (2004) A Synthetic Biohazard Non-proliferation Proposal. http://arep.med.harvard.edu/SBP/Church_Biohazard04c.htm.
- Conferees, SB2.0 (2006) Public draft of the declaration of the second international meeting on synthetic biology. <http://hdl.handle.net/1721.1/32982>.

- Cropp, T. A., Anderson, J. C. and Chin, J. W. (2007). Reprogramming the amino-acid substrate specificity of orthogonal aminoacyl-tRNA synthetases to expand the genetic code of eukaryotic cells. *Nat Protoc* 2, 2590-600.
- Fischer M., Maurer S.M. (2010) Harmonizing biosecurity oversight for gene synthesis. *Nature Biotechnology* Volume: 28, 20–22.
- Garfinkel M S, Endy D, Epstein GL, Friedmann RM (2007) Synthetic genomics: options for governance. <http://www.jcvi.org/cms/fileadmin/site/research/projects/synthetic-genomics-report/synthetic-genomics-report.pdf>.
- Herdewijn, P. and Marliere, P. (2009). Toward safe genetically modified organisms through the chemical diversification of nucleic acids. *Chem Biodivers* 6, 791-808.
- Implementation Support Unit (2008) Biosafety and biosecurity. UN document BWC/MSP/2008/MX/INF.1, Geneva. <http://daccessdds.un.org/doc/UNDOC/GEN/G08/618/92/PDF/G0861892.pdf?OpenElement>.
- Industry Association Synthetic Biology (2009) The IASB Code of Conduct for Best Practices in Gene Synthesis Cambridge, MA. Nov. 3, 2009. http://www.iasb.eu/tasks/sites/synthetic-biology/assets/File/pdf/iasb_code_of_conduct_final.pdf.
- International Gene Synthesis Consortium (2009). Harmonized screening protocol: gene sequence and customer screening to promote biosecurity. http://www.genesynthesis-consortium.org/Harmonized_screening_protocol.html.
- Jackson, RJ. et.al. (2001) Expression of mouse interleukin-4 by a recombinant ectromelia virus suppresses cytolytic lymphocyte responses and overcomes genetic resistance to mousepox, *Journal of Virology*, 75 pp.1205-1210.
- Kelle A. (2007) Synthetic biology and biosecurity awareness in Europe, Vienna, IDC. http://www.synbiosafe.eu/uploads///pdf/Synbiosafe-Biosecurity_awareness_in_Europe_Kelle.pdf.
- Kelle A. (2009) Ensuring the security of synthetic biology—towards a 5P governance strategy. *Systems and Synthetic Biology* 2009 December; 3(1-4): 85–90. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2759433/?log%24=activity>.
- Maurer, S., et al. (2009) Working paper: making commercial biology safer: what the gene synthesis industry has learned about screening customers and orders. http://gspp.berkeley.edu/iths/Maurer_IAsB_screening.pdf.
- National Research Council (2006) Globalization, biosecurity, and the future of the life sciences. Committee on Advances in Technology and the Prevention of Their Application to Next Generation Biowarfare Threats. The National Academies Press, Washington, DC.
- National Science Advisory Board for Biosecurity (2006). Addressing biosecurity concerns related to the synthesis of select agents. http://oba.od.nih.gov/biosecurity/pdf/Final_NSABB_Report_on_Synthetic_Genomics.pdf.
- National Science Advisory Board for Biosecurity (2010). Draft Report of the National Science Advisory Board for Biosecurity. [http://oba.od.nih.gov/biosecurity/pdf/NSABB%20SynBio%20DRAFT%20Report-FINAL%20\(2\)_5-21-10.pdf](http://oba.od.nih.gov/biosecurity/pdf/NSABB%20SynBio%20DRAFT%20Report-FINAL%20(2)_5-21-10.pdf).
- Rosengard AM (2002) Variola virus immune evasion design: expression of a highly efficient inhibitor of human complement', *Proceedings of the National Academy of Sciences USA*, 99 pp.8808-8813.
- Schmidt, M. (2010). Xenobiology: a new form of life as the ultimate biosafety tool. *BioEssays* 32, 322-31.

-
- Schmidt M, Pei L (2010) Synthetic Toxicology: Where engineering meets biology and toxicology. *Toxicological Sciences*. doi: 10.1093/toxsci/kfq339.
- Steinbruner J., Harris E.D., Gallagher N., Okutani S.M., (2007). Controlling Dangerous Pathogens. A Prototype Protective Oversight System, College Park: University of Maryland.
- U.S. Department of Health and Human services (2009) Screening framework guidance for synthetic double-stranded DNA providers. *Fed. Regist.* 74, 62319–62327 <http://edocket.access.gpo.gov/2009/e9-28328.htm>.
- Wadman, M. (2009) U.S. drafts guidelines to screen genes. *Nature News* (4 december 2009) <http://www.nature.com/news/2009/091204/full/news.2009.1117.html>.
- Wang, L., Xie, J. and Schultz, P. G. (2006). Expanding the genetic code. *Annu Rev Biophys Biomol Struct* 35, 225-49.
- Wang, Q., Parrish, A. R. and Wang, L. (2009). Expanding the genetic code for biological studies. *Chem Biol* 16, 323-36.
- World Health Organization (2004) *Laboratory Biosafety Manual, Third Edition*, Geneva.
- World Health Organization (2006) *Laboratory Biosecurity Guidance*. http://www.who.int/csr/resources/publications/biosafety/WHO_CDS_EPR_2006_6.pdf.
- 1540 Committee webpage (2010) <http://www.un.org/sc/1540/index.shtml>.