Analysis of the Threat of Genetically Modified Organisms for Biological Warfare

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EXECUTIVE SUMMARY

Evaluating the potential threats posed by advances in biotechnology, especially genetically modified organisms (GMOs) and synthetic biology remains a contentious issue. Some believe that, inevitably, these advances will lead to a catastrophic biological attack. Others believe that, despite these advances, the scientific and technical requirements, as well as the fundamental laws of natural selection will prevent such an attack.

To better understand this issue, this study narrowed the scope of consideration in several dimensions. First, our analysis primarily focused on what we defined as a “catastrophic biological attack”, with a required level of damage more associated with biological warfare than bioterrorism. This damage would need to be direct in nature where the effect is more physical than psychological. Second, this biological attack would be restricted to the United States, not another nation or entity. In this sense, U.S. geography, climatology, infrastructure and medical systems play to counterbalance any potential biological attack. Even within a more narrow scope, there remains inherent complexity and uncertainty which, combined with the considerable rate of change for biotechnology, defies a simple, straightforward answer.

We approached the issue by establishing an “Analytical Framework”—a baseline of the technical requirements to “play” in the field of GMOs at the scale of biological warfare. The primary focus of the framework are those aspects of the technology directly affecting humans by inducing virulent infectious disease, or through expression of toxins or suppression of the immune response of target subjects. Parallel threats exist for animals and plants in the food chain and, secondarily, in the ecosphere. Although not specifically included in this analysis, those threats can also be evaluated within the analytical framework. To establish our analytical framework, we focused on the engineering of novel single-cell microorganisms previously unknown in nature as described by four conditions:

1. Modification of known pathogen microorganisms to new functionalities
2. Modification of nonpathogens to become pathogenic
3. Synthesizing pathogenic microorganisms de novo
4. Synthesizing completely artificial or “abiotic” pathogenic “cells” or biomolecules.

We conclude that, broadly stated, peaceful scientific advances, global statistics and demographics of GMOs suggest that the potential for corruption of biotechnology to catastrophic malevolent use is considerable. At a more detailed level, we find that there are tangible opportunities for many potential adversaries to acquire, modify and then manufactures to scale a potential GMO pathogen. Further development of a modified pathogen for use in a full scale direct catastrophic biological attack is feasible, but the full spectrum of technologies for scale-up, testing, packaging, weapon production and employment will most likely require the resources of a nation state or comparably-resourced organization. We recommend that, in concert with “science-based” analysis, further efforts to expand and utilize this analytical framework be undertaken to better characterize the future threat from GMOs as well as other emerging threats such as those derived from systems or synthetic biology and bioregulators.

1 We define a catastrophic biowarfare attack as one having direct physical scale, such as the loss of a major U.S. city or national system; (As such, the 2001 U.S. Anthrax attacks would not qualify)
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Introduction

a. What is the issue?

Evaluating the potential threat posed by advances in biotechnology, especially genetically modified organisms (GMOs), and synthetic biology remains a contentious issue. The rapid development of the tools of molecular biology and metabolic engineering has enabled the development of chimeric organisms which possess characteristics which are not native to the wild variant. This is commonplace in the area of biomanufacturing, where genes are introduced into organisms such as *E. coli* and products manufactured via large-scale fermentation. More recently, entire metabolic pathways, albeit of limited complexity, have been engineered into organisms, for example, for the production of artemisinin in yeast. In addition to such metabolic engineering projects, whole genomes are being sequenced, leading to the possibility of creating organisms *de novo*.

Numerous lectures, briefings and articles have argued that the dual use nature of biotechnology, the training of foreign students in American universities, and the easy availability of information on the internet have given potential adversaries access to biological weapons of unimagined which pose an existential threat. Some believe that, inevitably, these advances will lead to a catastrophic biological attack.

Others have argued the opposite that making all information publicly available will enable a more universal “white biotechnology” which will ultimately monitor the field and provide the means to defeat any threat developed by adversaries. It has been argued that, despite these advances, the scientific and technical requirements, as well as the fundamental laws of natural selection, will prevent such an attack.

An example of the controversy is represented by statements such as that found on the web site of the Hastings Center, which states that,

“Research suggests that synthetic biology may soon be a technology of choice for a nation or terrorist hoping to develop or acquire a pathogen for use as a weapon”, however, without explicit supporting references.

To further demonstrate the depth of the issue, a brief listing of the current arguments For/Against the likelihood of a catastrophic biological attack being brought about by advances in synthetic biology follows.

Arguments FOR include:

- Advances in the science and technology of genetics, writ large.

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- Growth of commercial GMO activities.

- GMO knowledge base and its availability.

- Simplicity and availability of required low-cost materials and equipment.

- Human abuse of antibiotics and other practices which make populations more vulnerable to a GMO.

- Occurrences of pandemic disease derived from natural genetic evolution.

Arguments AGAINST include:

- Given the complexity of living organisms and their genetic makeup and responses, it is extremely difficult to predict the outcome of any genetic modification.

- The very limited success of “gene therapy” - peaceful medical objectives of genetics for new therapeutics and “individualized” gene based treatments are as yet unrealized.

- Nature is intolerant of modifications or new organisms and tends to select against them. Natural evolutionary processes make/break GMOs continually for the last three billion years, and it is unlikely that humans will outdo that.

- Extreme technical difficulties of “weaponization” for most potential GMO pathogens.

- An unimpressive history of bioterrorist attacks.

b. Complexity of the task.

The threat comprises an extremely diverse set of potential actors, tactical and strategic objectives, candidate targets to meet those objectives, candidate agent organisms appropriate to each, and a wide range of practical approaches for acquiring, modifying, and delivering threat organisms to their intended targets. Moreover, the science and technology of GMOs are, and will continue to be, a moving target. The field is expanding and knowledge and capabilities disseminating globally at a phenomenal rate. Today’s analysis may quickly be overcome by other developments in the near future.

c. Scope of Study and Deliverables.

This analysis focuses on the development of a robust and adaptable analytical framework for evaluating the threat posed by genetically-modified microorganisms, particularly those created using synthetic biology. The framework also addresses requirements for quantity production, packaging and delivery of threat agents to achieve a direct scale of damage against the U.S. at the level of biological warfare.
The analysis addresses a set of key questions, outlined in subparagraph d. below. Given the complexity of the task as described above, we do not present a full answer to these questions. Rather, the assessment attempts to lay out the essential technical requirements and alternative approaches available for developing a practical threat GMO. These form the point of departure for an analytical framework that takes into account the range of potential threat actors and objectives. These, together with the analysis of the threat development process, can be used to develop practical scenarios and evaluate the relative risks and benefits associated with different actors and objectives. Such a framework can be used to test various hypotheses and measure our depth of understanding.

d. Key Questions

1. What is the nature and scope of the threat, if any, posed by GMOs, to include the potential to develop completely de novo organisms or completely artificial abiotic systems?

2. What are the fundamental processes and global state of the art for creating GMOs?

3. Beyond the technical means to create a GMO, what might the follow-on requirements for “weaponization” include?

4. What are the capabilities and incentives for foreign states, transnational groups, small terrorist groups, or individuals to attempt to develop a significant GMO threat?
Background

The field of genetic modification of living organisms for human use has undergone explosive change. Since its first pragmatic elucidation in 1953, DNA structure and genetic engineering has extended its reach into agriculture, animal husbandry, medicine, and even organic materials. At its scientific limit, researchers are now applying genetic engineering to attempt to create completely new or de novo entities outside of the boundaries of normal organic reproduction or assembly. This section establishes our view of what a GMO is, provides examples of key and recent advances, and then characterizes the size of the effort (market) and its rate of change.

Multiple references and definitions of GMOs exist. For the purpose of this study, we approach the subject in its general form as described below.

A genetically modified organism is one whose genetic characteristics have been altered by the insertion of a modified gene or a gene from another organism using the techniques of genetic engineering. Genetically modified organisms encompass a wide spectrum of single and multicellular organisms, including plants and animals. This effort specifically addresses microorganisms (single cell biota and viruses). Organisms modified by insertion of genes from another organism are also referred to as “transgenic” organisms.

a. Historical and Recent examples of Genetic Engineering.

While the scope of this assessment focuses on single celled organisms, genetic modification of complex multicell organisms is a major commercial activity. To understand the current and future path of genetic engineering, it is useful to review some of the initial successes in this field, the various areas of application which have ensued, and then, as a subset, several key genetic engineering milestones relevant to biodefense.

Since the early 1990’s genetically engineered plants have been commercially available. So-called “first generation” transgenic plants have been engineered for characteristics that enhance the agricultural yield and marketing. Such characteristics include resistance to pests, herbicides and extreme climates, as well as improved product shelf life. For example, since their first commercial cultivation in 1996, plants have been genetically modified for tolerance to the herbicides glufosinate and glyphosate. A “second generation” of transgenic plants, now in research and development, is aimed at enhancing consumer satisfaction by enhancing taste,
texture, or appearance of produce. To date, no second generation transgenic plants are on the market.

Genetically modified/transgenic animals are used in a wide range of applications. Simple organisms such as fruit flies have been used to study the effects of genetic changes across generations. Transgenic mice are often used to study cellular and tissue-specific responses to disease.

Transgenic bovines and goats have also been developed to express a variety of useful biologically derived products. Among the first of these was “Herman the Bull”, who was genetically modified in 1990 with a human gene sequence while in embryonic form to produce lactoferrin, an immune system protein. This was followed by the development of a transgenic goat that expressed proteins for silk (similar to spider silk) developed by the Canadian firm, Nexia, under the trade name BioSteel. On February 6, 2009 the U.S. Food and Drug Administration approved the first human biological drug, also extracted from goat’s milk. The drug, ATryn, is an anticoagulant which reduces the probability of blood clots during surgery or childbirth.

Gene therapy, involving the use of viruses as a vector for introducing generic material into cells, has had some success in treating genetic disorders such as severe combined immunodeficiency, and treatments are being developed for a range of other currently incurable diseases such as cystic fibrosis, sickle cell anemia, and muscular dystrophy. Genes introduced in this manner are not transmitted to the next generation. Gene therapy targeting the reproductive cells—so-called “Germ line Gene Therapy”—at present carries an unquantifiable risk associated with interfering with other genes, hence near-term development and commercialization of this technology is unlikely.

In 2009, scientists in Japan announced that they had successfully transferred a gene into a primate species and produced a stable line of breeding transgenic primates for the first time.

Genetically modified bacteria have become commonly used as a means for producing large amounts of pure human proteins for use in medicine. Examples include production of insulin to treat diabetes, clotting factors to treat haemophilia, and human growth hormone to treat various forms of dwarfism. In addition, advances in biomedical research continue to build a growing store of knowledge directly applicable to the development of GMO threat agents. In recent years there have been a number of substantial development bearing on the potential threat of

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9 Erickson, Britt (10 February 2009). FDA Approves Drug From Transgenic Goat Milk.
10 American Journal of Law and Medicine, FDA Regulation –An Answer to the Questions of Human Cloning and Germline Gene Therapy, Boston University School of Law, 2001
11 Cyranowski, David, Marmoset Model takes Centre Stage, Nature, 459-523-527, May 2009
genetically modified or synthetically produced microorganisms. Genetic research in the field of biodefense relevant activities has also flourished. Several key examples follow.

- In 1981 scientists cloned a full-length virus genome (Poliovirus) that was infectious to mammalian cells and demonstrated the basis of the ability to replicate an infectious RNA virus.\(^\text{12}\) Significantly, in 2002, Cello et al. reported purely chemical synthesis of an infectious Poliovirus in the absence of any natural template.\(^\text{13}\)

- The potential for modifying organisms to significantly enhance virulence and mortality rate was shown when modified mousepox virus [in the same family (Poxviridae) as smallpox], intended for use as a contraceptive, proved 100% deadly by circumventing host immune defenses, even in previously immunized (vaccinated) animals.\(^\text{14}\)

- Genetic information even for highly virulent pathogens is widely available. The complete genome sequence of 45 variola strains providing supplemental material with gene organization of smallpox is freely available on the web.\(^\text{15}\)

- Cloning and recovery of infectious Ebola virus and of a mutant more cytotoxic than the natural wild-type.\(^\text{16}\)

- Later generation of a complete infectious genome (5,400 bases long in a bacteriophage) from synthetic oligonucleotides synthesized according only to the sequence reported in GenBank. The synthesis and assembly of this organism were completed in only 14 days and without a need for accessing any living organism.\(^\text{17}\)

- In 2008, Israeli researchers published a procedure for the \textit{de novo} construction of error-free DNA molecules from error-prone commercially available oligonucleotides.\(^\text{18}\) This ability was cited as having the potential to allow masking of an intended synthetic molecule or organism during purchase of oligonucleotides.

- In 2010, the J. Craig Venter Institute reported the successful synthesis of a complete microbe genome comprising over 1.0 million base pairs, and insertion of same into a microorganism capable of reproducing. However, assertions that this constitutes a fully-synthetic life form are arguably overstated, but the demonstrated ability to replicate the

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\(^\text{13}\) Paul Cello, E Wimmer, \textit{Chemical synthesis of poliovirus cDNA: Generation of infectious virus in the absence of natural template}. Science, 2002, 9 (297), No. 5583


\(^\text{15}\) J.J. Esposito et al., \textit{Genome sequence diversity and clues to the evolution of variola (smallpox) virus}. Science, 2006, 313, 807-812


\(^\text{18}\) G. Linshiz et al., \textit{Recursive construction of perfect DNA molecules from imperfect oligonucleotides}. Molecular systems biology 4: 191; doi:10.1038/msb.2008.26]
full DNA sequence is a substantial accomplishment. Hence, the claim would be better qualified as the assembly of a very simple life form by removal and addition of already living materials into a single cell structure.

b. How large is the GMO market and global enterprise?

Since the initial elucidation of the structure of DNA in 1953, and despite some market resistance (non-GMO attitudes, government and academic restrictions) the market for GMO products has rapidly expanded. Agricultural, chemical, pharmaceutical, industrial biotechnology (including energy) fields have all benefited from GMO activities and experienced levels of growth that outpaced other national employment. In 2009, commercial growth in the U.S. produced income of $75 billion dollars to a group of approximately 650 bioscience companies.

As an example, the global commercial value of biotechnology crops grown in 2008 was estimated to be $130 billion. The United States Department of Agriculture (USDA) reports on the total area of GMO varieties planted. According to the National Agricultural Statistics Service, the states published in these tables represent 81–86 percent of all corn planted area, 88–90 percent of all soybean planted area, and 81–93 percent of all upland cotton planted area (depending on the year).

Underlying this commercial growth are significant advances in the state of the art in genetic engineering and synthetic biology. The field has benefited from the confluence of several important technological trends: The discovery and subsequent rapid development of the field of genomics, the development of new tools and techniques for inspection and manipulation of matter at molecular levels, coupled with advances in information technology enabling efficient storage, processing, and dissemination of the vast amounts of data generated by advancing research in these areas.

The Number of DNA “synthesis foundries” world-wide has continued to grow. As of November 2010 there are an estimated thirty countries capable of synthesizing genes or genetic sequences of 1000 base pairs or larger. Including private, government and academic gene synthesis organizations a partial open source list yields: China (36), Germany (20), Great Britain (14), France (9), Russia (8), India (6), Canada (7), Netherlands (6), Israel (1), Iran, (1), and several others.

Private DNA “synthesis foundry” companies’ operations and reach are multinational. Eurofins MWG Operon has foundries in the US, Germany, and India; Takara Biotechnology (Dalian) in China is a subsidiary of Takara Bio Inc. of Japan; ThermoFischer of the US has a foundry in Lithuania; Origene operates foundries in both the US and China; and the Zelinsky

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19 U.S. Biosciences employment growth 2001-2008, from 100K to 600K Battelle, 2010
20 Battelle, 2010 Biotechnology Summary; Net Income 2009; dated 2010
22 USDA Economic Research Service, Adoption of Genetically Engineered Crops in the USA, Table http://www.ers.usda.gov/Data/BiotechCrops/
23 Defense Intelligence Agency, Unclassified Memorandum – NCI Response to ECBC, 9 November 2011
Institute Inc. of the US markets the products and capabilities of the Zelinsky Institute of Organic Chemistry in Russia.

The breadth of examples of key scientific breakthroughs described earlier offer evidence of scientific to commercial strength and public acceptance, and suggests that biological engineering is coming to be viewed in a manner similar to that of traditional engineering. The technical events and markers in the area of microorganisms demonstrate the practicality of manipulating of microorganisms and pathogens to change their characteristics. The advent of synthetic biology holds promise for novel and perhaps completely artificial, “abiotic”, functioning cells.

c. Rate of Change for Biotechnology/GMO technology

The rate of change for Biotechnology/GMO technology can be appreciated by considering job growth and the economics of GMO technologies. With respect to job growth in the U.S., between 2001 to 2008 employment in the bioscience fields rose from 100,000 jobs to over 600,000 (600%).

Considering the economics of GMO technologies, broad and growing markets for all types of GMOs, coupled with the widespread availability of basic information and technology, have driven rapid development and dissemination of the technology. The costs of both sequencing and synthesizing genetic material (key enabling capabilities for GMO development) have dropped dramatically in recent years (See figure 1.) and these trends are expected to continue. At present, costs for synthesis of short sequences of DNA are running as low as 0.3 Euros (40 cents/base pair), and costs for sequencing in 2010 are approaching $1.0/million base pairs. As a point of reference, there are some 2.9 billion base pairs in the haploid (chromosomal) human genome.

![The Carlson curve](http://www.synthesis.cc/)

Figure 1. “Carlson Curve”. http://www.synthesis.cc/

These include modifications based entirely on synthesized DNA where, as outlined above, the sequence length of purely artificial DNA has grown from a few thousand base pairs to over 1.0 million in the past decade.

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24 U.S. Biosciences employment growth 2001-2008, from 100K to 600K Battelle, 2010

This significant rate of change reflects the growing social awareness of global health issues and the increasing footprint of the pharmaceutical, medical, agricultural and industrial biotechnology sectors in national economies (not necessarily related). This has resulted in both rapid growth and globalization of genetic engineering capabilities. GMO advances to identify, characterize, modify, fabricate (clone, in-vitro replication, etc.), and stabilize DNA have all advanced rapidly and substantively. In this context, many of the arguments in favor of GMO threat expansion are understandable.
Technical Discussion: Scientific Principles Underlying GMOs

This section details the microorganisms of interest in this study, and then outlines the basics of genetic modification of microorganisms and the availability of various means for developing potential threat agents.

a. Microorganisms of interest

1. Bacteria. Bacteria are unicellular organisms. The unit of life is the cell, the smallest entity capable of displaying attributes associated with the living state, including growth, metabolism, stimulus response and replication. Bacteria can be modified by conducting modification of the cell while in a host organism, altered by chemical and/or organic means, or modified by plasmid introduction (circular DNA without DNA synthesis). Of the options available, plasmid introduction provides the most predictable product and outcomes and, by comparison, is inexpensive and technically trivial. However, stability of the plasmids over generations is not ensured.

2. Viruses. Although viruses are not technically alive, they represent supramolecular assemblies that act as parasites within host cells, underscoring the functional “culling out” of specific cellular processes, albeit within the confines of living cells. Viruses can be modified via DNA or RNA manipulation, where virulence factors can be spliced into the virus. Other modifications include changing the viral protein coat such that they can target specific type cells.

3. Listed but not included in Framework Analysis

   * Multi-cell pathogens
   * Toxins (Chemical products of living cells.)
   * Fungi (Robust organism; no genetic manipulation needed)
   * Prions (Generally not subject to genetic modification)

b. Options for acquiring pathogenic microorganisms for use as biological agents

Acquisition and use of naturally-occurring pathogens is a viable baseline for the development of a rudimentary biological agent. An example of a naturally-occurring organism that has been promoted as a biological agent both by nation states and terrorists is anthrax. The primary challenge is that acquisition of cultures “in-the-wild” requires visiting an area where the disease of interest is active. However, given an opportunity, the necessary samples can be collected and preserved by any adequately trained technician. Alternative methods, in general ascending order of difficulty include:

   * Obtaining an agent from a research center where work is being performed.
* Ordering from one or more culture collections maintained world-wide.

* Creating it either by modifying another pathogen or, in the case of viruses, synthesizing it from its obtainable components using conventional gene-splicing techniques or outsourcing to a DNA sequencing companies (also referred to as DNA foundries).

* As noted in the Background, RNA and DNA sequences for both viruses and microorganisms have been synthesized entirely from their genetic code.

* Finally, synthetic biology, including ongoing research in so-called “protocells”

The emergence of DNA foundries adds a new dimension to the potential threat. In the past, research activities have extracted and used gene splicing techniques to modify genetic materials, an approach which is labor intensive. DNA/Gene synthesis is being widely advertised as a more cost effective, less time-consuming approach.

Culture collections with pathogen stocks exist in many countries. General DNA materials (including possible pathogen stock) are normally developed or established as a unit “culture stock”, not as a single cell or bacterium. Culture stocks are, by standard, a test tube sized colony of the pathogen in a slant auger gel. The culture stock must be maintained at – 80 to -90 degrees F until it is needed for amplification, at which time it is thawed and grown in an appropriate medium, such as a 25-50 liter fermentation tank.

The World Federation for Culture Collections, World Data Center for Microorganisms database lists some 581 Culture Collections in 68 countries, holding over 1.6 million culture samples. In descending order, based on number of culture centers, the countries listed are Brazil (60 centers), Thailand, France, Australia, Japan, India, China, USA, Canada, the UK, Indonesia, Mexico, Russian Federation, and Republic of Korea (15). In terms of number of cultures maintained, the leaders (again in descending order) are: The US (210,276), Brazil, Japan, Denmark, United Kingdom, Netherlands, Australia, China, Republic of Korea, Canada, France, India, Belgium, Sweden, Germany, and the Russian Federation (45,655).

Most major academic institutions and national governments have some level of involvement in research. The Militarily Critical Technologies Program (MCTP) has estimated that there are over 400 locations around the world that maintain cell cultures that might be used as starting points for biological agent development, and biological materials from these culture collections are generally available to research centers world-wide. A significant consideration in terms of government oversight and security is fewer than half of the 581 collections are in government facilities.  

The statistical breakout is shown in Figure 2.

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c. Methods to modify or create a threat GMO

Synthetic biology holds potential for engineering pathogenic organisms not found in nature, either by introduction of synthetic genes into an existing natural organism, or by completely artificial “abiotic” synthesis of organisms. The potential effectiveness of microorganisms as biological agents can be enhanced in a number of diverse ways:

* Modify the organism by putting a molecule on the coat of an existing pathogen that causes it to bind more efficiently to the host target cell.

* Insert genetic material that encodes for a toxin (typically a protein). An example is the insertion of a plasmid with the 0157H gene in E. Coli. The genome of the cell isn’t altered, but it carries genetic material in the plasmids that encode the expression of toxins.

* Modify the genome of the microorganism itself to increase infectivity and virulence.

* Insert DNA sequences that bind to the host so that the host’s immune reaction is suppressed.

* Create a completely artificial or “abiotic” pathogen. Unlike current synthetic biological approaches, an abiotic organotypic approach would abstract the functionalities of living systems without copying their components.27

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27 Dr. James Valdes, Ph.D., “Transformational Countermeasures Technology Initiative”, RDECOM, 2008
The genetic material for modification may be either derived from natural organisms using standard recombinant DNA techniques, or produced by DNA synthesis, the latter being much less labor intensive. In recent work, DNA sequences on the order of 1 million base-pairs have been synthesized entirely from digitized genome sequence information, and the resulting organisms were phenotypical and capable of self-replication.\(^28\)

As is discussed in greater detail throughout this report, the tools and information required for genetic modification of microorganisms are readily available worldwide. The growth of synthetic biology, with its “engineering approach”, is expected to eventually lead to development of cheap, highly standardized building blocks (e.g., so-called Bio-Bricks\(^{TM}\)) and the design rules required for their functional assembly being widely disseminated.

d. Personnel and Costs

Assertions to the effect that a high school graduate can develop an effective biological weapon are arguably overstated. However, the work can be successfully accomplished by a small cadre; a trained clinician (if the feedstock is gathered in the wild), a graduate microbiologist, and a good laboratory technician are probably an adequate minimum staff. Depending on the nature of the intended attack, quantities of modified natural organisms sufficient to infect an individual, a small area or small concentrated group are within the capability of an appropriately trained individual, such as a competent microbiologist or medical clinician.

Development of novel (i.e., not known to be naturally-occurring) GMOs exhibiting unique designer characteristics requires substantially greater knowledge and capability. Many industrialized nations have laboratories capable of analyzing which immune response modifier genes in humans and livestock, when inserted into an organism together with pathogenicity (e.g., adherence and invasive) factor, will yield highly infectious pathogenic organisms.

In the development of biodefensive measures, however, the cost associated with characterizing genes of various species, including humans, that increase disease resistance or susceptibility is relatively high. There are approximately 25,000 genes in the human genome. The determination of the most preferred assembly of genes that will yield very high or very low susceptibility to infection will require significant financial commitments and fairly sophisticated research skills.

Offsetting the cost, results of research in this area is widely published and the resulting information stored in open-source repositories. The bioinformatic tools and techniques to access and manipulate the data are likewise widely shared globally.

e. Skills required to modify or create a threat GMO

The skills required to use a GMO as a threat falls into four broad categories. The first three, gene mapping, functional genomics, and bioinformatics, broadly comprise the discipline of genomics. The fourth category is, at the current state-of-the-art, less well developed. It comprises a loosely-

defined set of subspecialties such as pharmacogenomics, toxicology, immunology, biostatistics, epidemiology, and biochemistry. The common thread of these is the central role of understanding how organisms react to infection and to exposure to toxins, and broadly define the area now known as systems biology.

Information, particularly regarding naturally-occurring organisms and gene sequences, is freely accessible. As noted in previous studies the genetic information on a vast number of microorganisms as well as animals, plants, and humans is well structured, largely standardized, and accessible at www.ncbi.nih.gov/Genbank/ and other bioinformatics repositories. GenBank, with the DNA Data Bank of Japan, and the European Nucleotide Archive (formerly the European Molecular Biology Laboratory Nucleotide Sequence Database) participates in the International Nucleotide Sequence Database Collaboration, an cooperative effort to gather and disseminate nucleotide sequence and annotation and links for the three major data repositories.

The INSD Collaboration has a uniform policy of free and unrestricted access to all of the data records their databases contain. Scientists worldwide can access these records to plan experiments or publish any analysis or critique. No use restrictions or licensing requirements are included in any sequence data records, and no restrictions or licensing fees may be placed on the redistribution or use of the database by any party. This means that the sequences of many potential threat biological agents are freely available, and the information needed to synthesize or modify them is freely accessible. Anyone may access the computational tools to design genetically engineered organisms free of charge.

The latest update to the Biomedical Section of the Militarily Critical Technologies List also identifies the following countries as having large scale datasets related to genetic engineering: Australia, Germany, Italy, Japan, The Netherlands, Sweden, and the United Kingdom.

As of April 2010, the International Nucleotide Sequence Database repository of DNA sequences exceeded 100 gigabases. In comparison, the whole genome of Ebola virus is approximately 19,000 bases. The Web also provides free access to bioinformatics applications and software tools for analyzing genomic information. There is also a growing body of knowledge and data on proteomics, thus linking nucleic acid sequence information with the biological functions of proteins. These databases and tools characterize the function of specific genes with ever-increasing detail and fidelity, coupled with the ability to “mail order” sequences from a growing number of DNA sequencing companies world-wide provide a baseline capability for genetic engineering of microorganisms.

For example, the European Bioinformatics Institute [www.ebi.ac.uk] (EBI), part of the European Molecular Biology Laboratory (www.embl.org/), provides on-line access to a comprehensive range of tools for the field of bioinformatics (over 135 are currently listed). Online information available includes:

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29 Sagripanti, Jose-Luis, Ramsbotham, Alan J. ECBC – TR-666 – “Global Survey of Research and Capabilities in Genetically Engineered Organisms that could be used in Biological Warfare or Bioterrorism” Edgewood Chemical and Biological Center, December 2008.

30 Militarily Critical Technologies List, Section 4, Biomedical Technology, June 2009

31 http://www.insdc.org/documents/feature_table.html
* Similarity and Homology - the BLAST or Fasta programs can be used to look for sequence similarity and infer homology.

* Protein Functional Analysis - InterProScan can be used to search for motifs in a protein sequence of interest.

* Proteomic Services NEW - UniProt DAS server allows researchers to show their research results in the context of UniProtKB/Swiss-Prot annotation.

* Sequence Analysis - ClustalW a sequence alignment tool.

* Structural Analysis - MSDfold or DALI can be used to query any protein structure and compare it to those in the Protein Data Bank (PDB).

* Web Services - provide programmatic access to the various databases and retrieval/analysis services.

* Tools Miscellaneous - Expression Profiler a set of tools for clustering, analyses, and visualization of gene expression and other genomic data.

Answers to most technical questions that may arise can be found on the World Wide Web at one of the help sites from the many universities that carry related activities within newly formed departments of bioinformatics or specific informatics resources. As the global knowledge base characterizing functional genomics and proteomics expands and is dispersed to less developed nations, it will be relatively easier and inexpensive to generate genetic combinations that will markedly increase infectivity and pathogenicity of any given organism.  

**f. Likely technical objectives of modifying or creating a threat GMO.** There are a number of objectives, each with their own technical requirements and technical difficulties that a perpetrator might pursue. These include, but are not limited to, either uniquely or in combination:

* Increase infectivity.

* Increase virulence/mortality rate.

* Diminish host immunity or confer antibiotic resistance.

* Increase survivability outside the host (i.e., environmental stability).

* Circumvent/degrade detection/protection measures.

A key observation however, is that the first several bullets drive a trade-off with regard to the quantity of material needed to pose a threat. Threats with a very high virulence and mortality

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32 MCTL Section 4.1 Host Genome Material in Virus-like Agent to Affect Soldier Capability, June 2009
rate, while psychologically devastating, tend to be self-limiting unless they also have high person-to-person infection rates, in which case they may take a while to “burn out.” In practice, the objectives of genetically-modifying organisms for use as biological agents will be driven by the user’s perceived threat/benefit analysis, difficulty of production, and the threat characteristics that they possess.

g. Catastrophic Attack and Weaponization

1. Weaponization involves co-opting the advances of genomics for malevolent purposes. For the objectives of this study, deployment of such weapons might result in catastrophic consequences. Further, we define a catastrophic attack on the basis that there is a direct scale consequence of the attack. This is unlike a typical terrorist attack where the main effect is psychological. For example, we do not consider the 2001 U.S. Anthrax Attacks to be “catastrophic”. Although there were significant indirect consequences, the actual numbers of people killed were very few. We would define a significant direct attack as one which results in thousands of casualties up to the loss of population equivalent to that of a major U.S. city.

Weaponization is the most difficult task in conducting biowarfare. Although it may be possible to create or modify a pathogen in a laboratory, the next steps of producing sufficient quantities of the pathogen, deriving a means to take it out of the lab and have it survive to the point of attack, and then to disseminate it successfully all pose significant challenges. Within those steps there is requirement for a means of delivery which is timely, sufficiently broad and with an effective “uptake” or infectivity in transmission. Weaponization techniques can vary greatly. As examples, one threat pathogen may require scale production, stabilization via inert coating for “encapsulation” and then aerosol distribution via mechanical means; another less controlled approach would be through contagious vectors, infecting and then releasing carriers of the pathogen to naturally replicate and deliver the threat. This section covers principles which will be further discussed in the next section on Framework of Analysis.

2. What does it take to produce a volume of pathogens to the scale needed for a significant or catastrophic attack? Depending on the type of pathogen, the amount needed for a catastrophic attack could range from several milliliters to 55 gallon drums. The respiratory doses for various microorganisms for an infection in humans (measured in ugs) range from 0.00000021 for Q-fever to .008 for anthrax. Respiratory doses for biotoxins such as Staphylococcal Enterotoxin B (SEB) (0.025 ugs) and Botulinum neurotoxic (4.5 ugs) are orders of magnitude higher. For comparison, the effective dose for the Nerve Agent VX is 70ug; many orders of magnitude greater than that of Q-Fever. 33

3. Materials. The materials required such as reagents, culture media and host vectors are readily available worldwide. These are all used in a variety of life sciences and environmental applications, and there are no effective restrictions on a potential enemy’s access to them.

33 Dr. Robert Armstrong, Dr. David Franz, Dr. James Valdes, *Bill Patrick’s Relative Aerosol Potency Chart, Biological Agents: Threat, Preparedness, Response and Myths*, presentation to European Commission, February 2009
4. Facilities. The cost of a facility for modifying, culturing, and replicating GMOs in quantity sufficient to pose a significant biological agent threat has been estimated by independent studies to be on the order of $200K to $250K. With such a facility and using proper scale-up bioprocessing techniques, one can amplify the volume of a test tube culture sample to a 25 – 50 liter basis within 24 hours.  

Table 1. Money * Genetic Engineering Comparative Costs

Compared to other projects that might be undertaken by governments or private organizations, the cost of equipping and staffing a laboratory scale bioprocessing facility, as shown in Table 2 below are trivial.

Table 2. Comparative Costs for a scale bioprocessing facility

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5. Ability to stabilize pathogen for delivery. Assuming an adversary is capable of accomplishing genetic modification and scale production of a threat pathogen, outside of an infectious disease model, the agent must then be prepared to be used as a weapon. Given the ambient conditions of sunlight, temperature, and exposure to other meteorological factors, most microorganisms do not survive unless specially prepared. The next critical step is therefore to stabilize the pathogen in a form that allows for such survival. Previous techniques included “microencapsulation”, in which the pathogen is coated with a protective material as, for example, in the coating of enzymes for laundry detergent; embedding in biofilms; and, in rare cases, use of living vectors in a manner similar to, but more controllable than, an infectious disease model.

6. Means of Delivery. Finally, to launch a catastrophic attack, the perpetrator must have some means of delivery. Possible methods include direct means of delivery via aerosol, indirect means via packaging and leveraging of US delivery systems (e.g., FEDEX, US grocery distribution system, Postal Service) and delivery via natural vectors.

h. Effectiveness of the tools of synthetic biology for threat GMOs.

As one respected scientist summarized: “Today, anyone with a high school education can use widely available protocols and prepackaged kits to modify the sequence of a gene or replace genes within a microorganism; one can also purchase small, disposable, self-contained bioreactors for propagating viruses and microorganisms. Such advances continue to lower the barriers to biologic-weapons development.”

Is this really true and, if so, how far is the barrier to biological weapons development being lowered? Being able to modify genetic material is one thing; understanding the end effects in terms of how such modification will affect the characteristics of the organism and its effects on a host organism’s physiology is something very different. There is strong evidence to suggest that intentional modification of a pathogen remains difficult.

From the point of view of the potential perpetrator, the challenge is to reliably predict the overall effects of the changes. For example, the intent of the Australian researchers in modifying mouse pox was to produce a contraceptive effect, and the subsequent lethality of the modified virus was an unintended side effect. Conversely a sequence of genetic material that codes for expression of a particular toxic protein may inadvertently suppress other functions essential to the reproduction or survivability of the microorganisms.

Arguments against GMO weapons include the limited success of “gene therapy”. During its inception, pursuant to the derivation of the human genome and advanced bioinformatics and DNA processing, it was posited that unique individual genomes would be determined and then used to prescribe gene-based therapeutics for a variety of diseases. With a few general exceptions, individualized gene therapies have not yet emerged. This lack of success is generally attributed to the observation that human genomics does not imply a specific one-to-one mapping of particular genes to singular specific health responses. Instead, the human gene composition

includes numerous redundancies where multiple genes or a system of backup genes can all play a role in immunity and response to a pathogenic challenge. Organisms and their genetic composition and host-pathogen interactions are exceedingly complex.

As a further example, analysis of one of the simplest pathogens, the prion, was conducted utilizing the latest methods of systems biology. Using multiple mouse models, gene expression data, and techniques such as subtractive biology, an initial set of more than 7,400 genes whose expression changed in response to prion infection was winnowed down to 333 which were critically involved in disease progression, and specific multiple effects on metabolic pathways were determined. 36 Such a systems biology approach could eventually lead to very targeted medical countermeasures, either prophylactic or therapeutic, but could also be used to predict or target the effects of a pathogen.

With respect to the de novo design of entirely new GMOs, there are additional challenges based on the spatial architecture and geometry of the cellular environment. 37 In this case, the intricacies of having cellular structures and processes come together in the correct spatial/temporal points to achieve proper function is an exceptionally difficult challenge. Building a synthetic cell, or even making a drastic modification to an existing cell, must account for this architecture.

Finally, to date, the results of previous biological attacks have been most unimpressive. The most recent instance was the 2001 anthrax mailings where, despite the perpetrator using a weapons grade pathogen and using the U.S. mail system for physical delivery of the anthrax spores, the results of over one billion doses mailed was five deaths. 38

In summary, developing a pathogen suitable for use as a biological weapon agent and its subsequent “weaponization” is not simple. Acquisition or creation of a pathogen, subsequent genetic modifications, amplification of the pathogen stock to a volume that is subsequently stabilized and delivered in a naturally hostile environment against a well-defended public accumulates many challenges.

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36 Leroy Hood, Finding Early Signs of Mad Cow Disease, Molecular Systems Biology, March 2009
37 Ochman and Raghavan, “Excavating the Functional Landscape of Bacterial Cells; Science 27 November 2009
38 The Unimpressive History of Bioterrorist Attacks, slide 21 Biological Agents: Threat Preparedness, Response and Myths
Framework of Analysis

a. Why a framework and for what is it used?

Why? Given the complexity and uncertainty of this issue, one cannot leap to a single, or even a multiple set of answers. A framework serves as a functional alternative to structure our thinking and understanding of the issues and ultimately serves as a platform from which to develop particular answers. The framework reflects the key dimensions of consideration which are used to “hang” ideas or facts on, test various hypotheses and measure the depth of our understanding.

What? Framework consists of key questions, subordinate metrics and facts needed to answer those questions.

The framework emphasis is on single cell organisms, viruses, and a limited group of multi-cellular fungal organisms that pose potential threats (such as those causing plant rust.) The framework developed includes placeholders for future elaboration on the threats posed by such organisms to multi-cellular life forms. As our understanding of the functioning of different metabolic pathways and the damaging effects of their products (such as proteins) improves, these placeholders can be expanded to assess whether the increased utility of GMOs relative to naturally-occurring organisms is likely to provide an incentive for their development.

b. Framework structure.

For the purpose of this study, our Analytical Framework is predicated by the current set of Technical Facts and Requirements to create and expand GMOs from single culture to scale distribution. Using that set of preliminary facts we then constructed a two dimensional framework to reflect the process chain necessary to conduct a deliberate scale bio-attack with a GMO, and then to assess the more subjective elements of “outcomes” as a function of “feasibility effects” and “cost-benefit” analysis The technical predicate and framework are depicted in the following graphics:
At the “meta level” these include 6 parametric variables:

1. Who are the likely perpetrators?
2. What it takes to make a single GMO culture stock?
3. What it takes to make scale quantities for a catastrophic attack?

What is takes to “weaponize” a GMO?
4. Embed pathogen
5. Means of delivery / dissemination
6. What are the potential GMO outcomes (Feasible Effect & cost/Benefit)?

Beneath each of these variables are additional subordinate variables, further amplifying the possible permutations and combinations of approaches and outcomes.

1. Who are the likely perpetrators?
   * “Grad student” accident or experiment
   * Lone terrorist
   * Disgruntled employee
2. What does it take to acquire one threat GMO pathogen culture stock? Summarizing the principle methods from Section III, Technical Facts:

Selection and genetic modification of organisms to create an initial pathogen stock can be done with relative ease. Alternative approaches to acquiring an initial stock are:

* Harvest from nature. By going to the origins of a pathogen, with the aid of a clinician, one can harvest “wild type” pathogens that are endemic to certain regions.

* Obtain it from a research center where work is being performed.

* Create it either modifying another pathogen or synthesizing it from its obtainable components using conventional gene-splicing techniques either in available third-party facilities or own dedicated facilities. This includes, at the current state-of-the art, outsourcing to DNA sequencing companies (also referred to as DNA foundries).

* By synthetic biological or abiotic techniques.

A significant complication is the likelihood that a nation state may mask its actions and involvement in a given attack by equipping surrogates such as individuals or small terrorist groups with the means to carry out a more sophisticated attack, as Iran is thought to do with conventional weapons and Hezbollah.

3. What does it take to embed the pathogen in a medium suitable for storage and dissemination without killing or disabling would-be attackers and without attracting the attention of police and intelligence agencies?

4. What are the means of delivery/dissemination?
   1. Dispersion (aerosol; FEDEX, grocery deliver system, etc.)
   2. Infection
   3. Poisoning
   4. Vectors (fleas, ticks, etc) “Reservoir Vectors” that give persistence – food, sewage, but infrequently humans

5. What are the potential GMO threat outcomes (Feasible Effect & cost-Benefit)?
* Humans. Induce large numbers of victims (setting aside the psychological effect, we can postulate “at rates of infectivity and virulence exceeding naturally-occurring pathogens as a pragmatic metric);

* Burdening health care systems / other economic burdens

* Agricultural and animal industries / Disrupt Food Chain (nutrition / Economic)

* Area denial / Disrupt operation of critical infrastructure

* Equipment contamination
Limited Current Analysis

The Analytical Framework captures the complexity and uncertainty of a potential GMO threat. From a probability and statistics viewpoint, the combinations and permutations available in the model yield approximately 300,000 possible pathways and outcomes. Researching each pathway and outcome with full enumeration is possible, but probably not useful, and a better use of the framework would be to demonstrate and evaluate both historical and proposed attacks from literature and intelligence estimates. Nonetheless, from the effort supporting the development of this limited framing and analysis, the following views are offered.
Preliminary Findings

Using the original set of research questions posed, we found:

1. Primary question: What is the nature and scope of the threat, if any, posed by GMOs, to include the potential to develop completely de novo organisms or completely artificial abiotic systems?

   * The likelihood of a completely artificial or abiotic single cell entity, much less a deliberate pathogen, is very small. To date, despite some published claims of an artificial life-form, biological science is, at most, still only emulating the otherwise natural fabrication of living entities.

   * Modification of existing pathogens to avoid detection, be more virulent or better weaponized is more likely, but probably only in the hands of nation-state or above level. Overall, the overhead to create/use GMOs as a military weapon is only plausible at nation-state or above level.

2. What are the fundamental processes and global state of the art for creating GMOs? The fundamental processes for creating GMOs are reflected in the Analytical Framework. The global state of the art for creating GMOs is more complex, but generally due to the significant global increases in the field of biotechnology, the primary capabilities to at least create one GMO culture is widely available.

3. Beyond the technical means to create a GMO, what might the follow-on requirements for “weaponization” include?

   * Outside the laboratory, nature tends to side with the defender since ambient conditions tend to kill or reduce effectiveness of GMOs. Evolutionary processes suppress man-made efforts to propagate pandemic like weapons. Nonetheless, as in nature, exceptions occur. Sunlight (UV); heat, cold, lack of availability of a suitable host organism, all comes into play; therefore:

     * The ability of most perpetrators to manufacture scale quantities (nominally 25 gallons) is apparent. However the final steps of pathogen stabilization and delivery will elude all but the very competent nation state adversary.

4. What are the capabilities and incentives for foreign states, transnational groups, small terrorist groups, or individuals to attempt to develop a significant GMO threat?

   * Although possession of a capability to develop a GMO threat is plausible by a non-nation actor, other than using GMO to avoid detection, there is no real advantage to do so.
and mounting a “catastrophic” pathogen attack is more easily accomplished without GMO overhead and uncertainties.

* The classified annex to this report includes a more detailed answer to this question.

Other relevant findings include:

* There is a trade space for some pathogens where increased virulence will result in “burn-out” within a confined geographical area, that is, those susceptible to the pathogen will succumb quickly, while those who aren’t will be immune. The propagation of the pathogen will then cease unless individuals break out of the confined area and further communicate the disease to new areas.

* Identifying and preventing any GMO attack will be problematic. Unlike other classes of weapons (e.g., nuclear devices, artillery pieces, etc.,) the science, technology, means of production and delivery of GMOs are demonstrably dual use. The path necessary to produce a beneficial GMO for commerce is often indistinguishable from that necessary to create something malevolent, and the path from a beneficial to a threat GMO is short and swift. The GMO threat generally cannot be detected by the normal intelligence collection and analysis methods.
Conclusion

We conclude that, broadly stated, peaceful scientific advances, global statistics and demographics of GMOs suggest that the potential for corruption of biotechnology to catastrophic malevolent use is considerable. At a more detailed level, we find that there are tangible opportunities for many potential adversaries to acquire, modify and then manufacture to scale a potential GMO pathogen. Further development of a modified pathogen for use in a full scale direct catastrophic biological attack is feasible, but the full spectrum of technologies for scale-up, testing, packaging, weapon production and employment will most likely require the resources of a nation state or comparably-resourced organization. We recommend that, in concert with “science-based “ analysis, further efforts to expand and utilize this analytical framework be undertaken to better characterize the future threat from GMOs as well as other emerging approaches and entities such as those derived from systems or synthetic biology and bioregulators.
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Annex A. Terms and Definitions

**Fundamental Genomics** – where research is generally directed towards the basic structure of human and microbial systems. Understanding the basic structure, organization, and function of human and microbial genomics potentially allows the development of genetically modified threat organisms resistant or immune to the natural defenses of the human body.

**Functional Genomics** – Once the structure of a genome is determined (sequenced), a major task remaining is to determine the function of each of the genes.

**Proteomics** – The complex set of proteins encoded by the cell during its lifetime is referred to as the proteome. One goal of genomics is to understand in detail, how the different genes encode for the synthesis (also known as expression) and assembly of proteins. For example, controlling the protein expression of antigenic proteins in a microorganism could make such a microorganism resistant to vaccines or undetectable by antibody-based diagnostics. Also, GMOs could be tailored to either express proteins or metabolize products that would be toxic to the host.

The underlying technologies for developing and exploiting genomic information can be divided into broad areas that support the three sub-fields of genomics described above.

**Gene Sequencing** – the basic “pick and shovel” work of determining the physical structure of the genome. The rate of gene sequencing and number of entities sequences has undergone hyperbolic expansion in the last ten years.

**Molecular biology and chemistry** – including technologies for rapid screening and combinatorial chemistry, are essential to advancing functional genomics, and to understand the biological and chemical effects of specific proteins and other biochemicals expressed during biological processes.

**Protein engineering and bioprocess engineering** extend molecular and genetic knowledge to optimize large scale and affordable production of organisms and biological materials.

**Bioinformatics** is a critical field comprised of state-of-the-art and entirely new information processing capabilities which are required to make effective use of the volume of data produced by biomedical research. Bioinformaticians can identify metabolic pathways from specific gene to end product, molecular structure, and even correlate gene sequences of threat viruses to the disease that they may cause. The informatics tools for systems biology, including databases such as Genbank and the Kyoto Encyclopedia of Genes and Genomes and analytical tools such as Cytoscape, now make possible the ready visualization and analysis of virtually any set of pathways, natural or contrived.

Definitions exist. What they all have in common, however, is that they see synthetic biology as the design and construction of new biological functions and systems not found in nature. More

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39 Jose-Luis Sagripanti, Alan J. Ramsbotham, Jr. “Global Survey of Research and Capabilities in Genetically Engineered Organisms that could be used in biological Warfare of Bioterrorism”, December 2008
precisely, synthetic biology seeks to mimic living systems by re-designing cells and tissues. Such approaches are bound by the limitations of cellular biology. As an example, genes, proteins, and functionalities are becoming increasingly fungible real-world entities that are being engineered as synthetic biology ‘parts,’ such as BioBricks™. “Synthetic biology may be especially powerful in this respect because it frees the design of biological systems for the process of natural evolution. The ability to sequence and then synthesize DNA (and even to invent new base code) adds a new layer to the power of nature: giving Synthetic Biology is a new area of biological research that combines science and engineering and encompasses a variety of different approaches, methodologies and disciplines, and many different humans the ability to design and redesign the biological systems of which they themselves are part.”

**Systems Biology** Systems biology is the integration of the many different levels of knowledge (genomics, proteomics, metabolomics) about cells and organisms to gain a global understanding of function. Most recently, the term synthetic biology has been adopted by an engineering cohort to define the process by which natural biological molecules (enzymes, DNA, proteins, etc.) are extracted from living systems and defined as basic building blocks to be reassembled in unnatural order and environments to create novel “devices or machines” that perform specific, predictable functions which may or may not be found in natural biological systems. This engineering approach differs significantly from “systems biology”, in that the individual biological constructs most suited to constructing a device are those units that act independently in contributing to the whole: the whole can be predicted from the sum of its individual parts.

**Abiotic cells** include “organotypic” approaches which abstract the functionalities of living systems without copying their components.

**Bioregulators** include a variety of neuropeptides which mediate many biological functions such as reproduction, metabolism, growth, temperature, heart rate, behavior, memory and emotional state. Examples include endorphins, enkephalins, and tachykinins.

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40 Wikipedia, general description of Synthetic Biology for laymen.
43 James Valdes, Ph.D “ANTS-TCTI” White Paper,
44 James Valdes, Jr. Ph.D., TCTI, RDECOM proposal, 2008
45 Norbert Herzog, Ph.D, Biotechnology, biodefense and nanotechnology Advances in Academia and Industry, Department of Pathology, University of Texas Medical Branch, TX
Annex B Recommendations

1. Investments. Given the findings from the overall study analysis and framework, it would be difficult/inappropriate to make specific investment recommendations in any particular field of technology or application. If there is a need for an investment determination, then in conjunction with a well defined set of assumptions and conditions, the framework should be utilized to make better informed decisions.

2. We recommend that the basis of future GMO or related bioware studies be shifted to a more science-based approach. In addition to the overall complexity, uncertainty and speed of change with respect to biotechnology, there are significant impediments with the efficacy of traditional threat analysis when used in this field.

3. In particular, standard methods of intelligence collection and analysis appear to be mostly based on literature and spoken intent, rather than technical competencies. Granted, there are fundamental difficulties to discern malevolent capabilities from the inherent dual-use nature of biotechnical development. Still, shifting towards a more science-based analysis, such as use of proxy technical means to test for key biowarfare processes, may better illuminate what the real threat is.

4. The analytical framework should be expanded to include the next “horizon” of potential biologically based threats, such as bio-regulators or potential threats brought about through systems biology. An additional area on new interest would include the recent development of microorganisms which can attack materials, such as silicon, which may produce a new category of “executable” biowarfare.

5. The results of any specific application of the frameworks towards a particular threat or context should be used to help update and inform the intelligence community. There is some strong potential that better warnings, indicators and collection methods might benefit from such analysis.