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The Impact of Synthetic Biology

Gi Na Lee and Jonguk Na*

Korean Minjok Leadership Academy, 600 Bongwha-ro, Anheung-myeon, Hoengseong-gun, Gangwon-do 225-823, Republic of Korea

ABSTRACT: Synthetic biology has recently been at the center of the world's attention as a new scientific and engineering discipline. It allows us to design and construct finely controllable metabolic and regulatory pathways, circuits, and networks, as well as create new enzymes, pathways, and even whole cells. With this great power of synthetic biology, we can develop new organisms that can efficiently produce new drugs to benefit human healthcare and superperforming microorganisms capable of producing chemicals, fuels, and materials from renewable biomass, without the use of fossil oil. Based on several successful examples reported, this commentary aims at peeking into the potential of synthetic biology.

In February 2012, the World Economic Forum (WEF) announced the "Top 10 Emerging Technologies". In first place was "informatics", due to the increasing importance of big data and social network services and rapidly advancing smart devices.[1] In second place was "synthetic biology and metabolic engineering".[1] The Global Agenda Council on Emerging Technologies described its reason for placing synthetic biology and metabolic engineering in second place on the WEF's ForumBlog: "*The natural world is a testament to the vast potential inherent in the genetic code at the core of all living organisms. Rapid advances in synthetic biology and metabolic engineering are allowing biologists and engineers to tap into this potential in unprecedented ways, enabling the development of new biological processes and organisms that are designed to serve specific purposes—whether converting biomass to chemicals, fuels, and materials, producing new therapeutic drugs, or protecting the body against harm.*"[2]

Synthetic Biology. Synthetic biology is changing the paradigm of biology and biotechnology. It allows the design and construction of new biological parts, modules, devices, chassis, and systems, in addition to reengineering cellular components and machineries that nature has provided.[3,4] For example, two seminal papers presenting the first synthetic gene networks appeared in 2000: an artificial toggle switch developed using a feedback system made of two cross-repressing genes[5] and a synthetic oscillatory network using three transcriptional repressors.[6] Scientists have even created "living photographs" made of bacteria, engineered on the basis of the design principles of synthetic biology. In short, *E. coli* was engineered by introducing genes that result in the formation of black pigment only when cells are in the dark. When a sheet of engineered *E. coli* is exposed to light passing through a slide with a designed photo image, only the parts exposed to light show the natural yellowish *E. coli* color while the rest turns to black.[7] During the past decade, numerous genetic circuits have been constructed by synthetic biology. These genetic circuits are in many ways analogous to electronic circuits. For example, Christopher Voigt and colleagues have developed a synthetic genetic edge detection program by engineering bacteria that identify the light-dark boundaries within a projected image of light.[8] Cells were made to produce, in the dark, a chemical that diffuses into the light regions. Then, cells were further engineered to produce a positive output only when they sense light and the chemical signal.[8]

Unlike synthetic biology, metabolic engineering, which aims at the purposeful modification of metabolic, gene regulatory, and signaling networks to endow cells with improved properties and performance, has been around for more than two decades.[9] Many successful examples of the enhanced production of chemicals, fuels, materials, and drugs through metabolic engineering have been reported, and more are expected to appear. While metabolic engineering has mostly been focusing on modification, redesign, and optimization of the existing cellular metabolism and regulatory circuits, synthetic biology provides additional tools through the synthesis of new enzymes, regulatory proteins, metabolic pathways, regulatory circuits, and importantly, very fine control of the local and global cellular networks.[3,4,10]

Microbial Cell Factories. After the advent of synthetic biology, microbial cell factories for the efficient production of chemicals, fuels, and materials have been more actively developed through the integration of metabolic engineering and synthetic biology.[11] Beyond traditionally sought biofuels such as ethanol, advanced biofuels such as butanol, hydrocarbons, and even new terpene-based biofuels are coming into view at improved production rates.[12] For example, butanol has much better fuel properties compared to ethanol but could not be efficiently produced until recently. Using the most popular bacterium *Clostridium acetobutylicum* for butanol production, system-wide analyses of metabolic pathways followed by metabolic engineering, including the introduction of a synthetic enzyme, resulted in an engineered *C. acetobutylicum* capable of producing butanol at high efficiency.[13] It was notable that redesigning pathway operation by attenuating the traditional cold channel (acidogenesis followed by solventogenesis) and reinforcing the hot channel (direct butanol forming pathway) allowed significant increase of butanol yield and selectivity.[13] Development of microbial strains capable of producing bisabolene, a terpene-based advanced fuel, is also an amazing example of what synthetic biology and metabolic engineering can do.[12]

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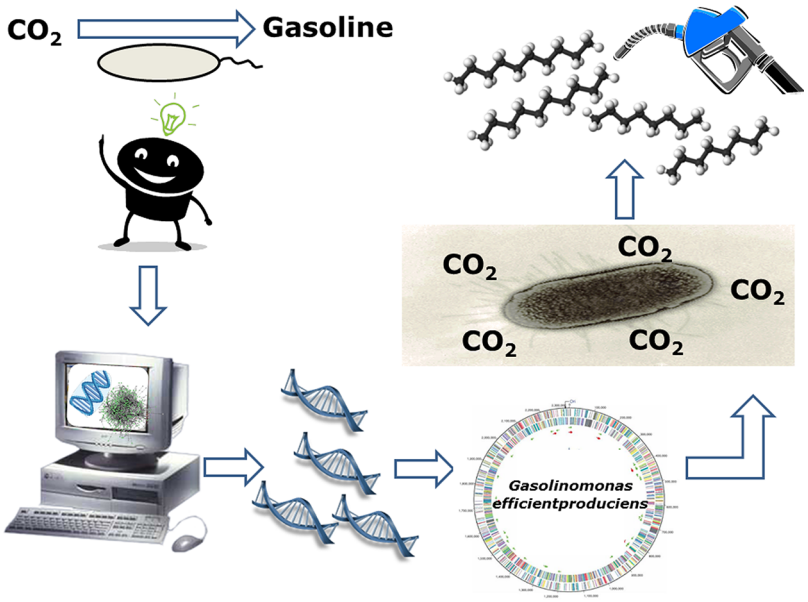

Figure 1. A procedure for developing an imaginary artificial microorganism, "*Gasolinomonas efficientproduciens*", capable of overproducing gasoline from carbon dioxide. One can generate an idea based on the necessary metabolic and gene regulatory pathways for directly converting carbon dioxide to gasoline, a mixture of C4−C12 hydrocarbons. The genome can be designed and optimized on a computer, which will be followed by its rapid synthesis and assembly. A robust chassis microorganism can be chosen, into which the synthesized genome is introduced. The artificial microorganism *Gasolinomonas efficientproduciens* is cultivated in a large-scale fermenter and overproduces gasoline directly from carbon dioxide.

On September 23, 2009, CNN reported on the making of plastics without fossil oil.[14] The plastic was polylactic acid (PLA), a biobased polymer that can be used in a wide range of applications, just like fossil oil derived polyesters. PLA is a non-natural polymer and has been produced through a two-step process: fermentative production of lactic acid, followed by a complicated chemical polymerization process. For the one-step biosynthesis of PLA in *Escherichia coli*, researchers combined metabolic engineering with synthetic biology to design a new non-natural metabolic pathway.[15,16] Creation of an enzyme converting *in vivo* generated lactate to lactyl-CoA and another enzyme that can polymerize lactyl-CoA into a lactate-containing polyester were the key steps in developing this engineered strain.[15] System-wide metabolic engineering was then carried out to enhance the production of PLA.[16] This approach can be extended to the biobased production of other chemicals and materials currently derived from fossil resources.[10−12]

In addition, more efficient production of existing drugs and synthesis of new drugs are possible through synthetic biology. Engineered microorganisms have been employed for the production of precursors of complex drug molecules such as the antimalarial drug artemisinin[17] and the anticancer drug taxol.[18] These examples demonstrate how synthetic biology contributes to establishing an environmentally friendly and sustainable chemical production system, as well as improving human healthcare.

Artificial Organisms. In 2010, a milestone paper on the creation of the first artificial bacterium was published. Researchers at the J. Craig Venter Institute had created a synthetic cell that can survive and reproduce itself relying on an artificial DNA sequence designed.[19] The genome of a small bacterium *Mycoplasma capricolum* was completely rewritten, and its DNA fragments were synthesized and assembled. The synthetic genome was transplanted into an empty *Mycoplasma mycoides*, which resulted in the creation of a new species named *Mycoplasma mycoides* JCVI-syn1.0.[19] Although *Mycoplasma mycoides* JCVI-syn1.0 is still far from a truly created cell, this work showed the possibility of creating an artificial organism for the first time.

More to Come. Synthetic biology is a great scientific discipline, allowing us to better understand the biological system and to optimally design biological parts, modules, and systems. As described above for the production of chemicals, fuels, materials, and drugs, synthetic biology can provide us with many benefits that cannot be solely provided by nature. Its integration with metabolic engineering allows the development of biological systems that can benefit humans and nature in many ways, from the creation of new drugs to the efficient production of renewable chemicals and materials.

What lies in the future? As the first artificial cell has been created, although mostly copied from what nature provided, it is natural to predict that more sophisticated metabolic, gene regulatory, and signaling networks of high performance can be completely designed, synthesized, and used to create a designer cell for the benefits of humans and the environment. Rapid advances in our knowledge of natural biological components and networks will allow us to better design optimally performing cells. Also, numerous functional gene modules and clusters can be newly designed or refactored based on what nature provided us with. A good example is refactoring the complete nitrogen fixation gene cluster and transferring it from one bacterium to another. The *Klebsiella oxytoca* nitrogen fixation pathway for converting atmospheric nitrogen to ammonia was refactored for better balanced expression and was transferred to *E. coli*, a non-nitrogen-fixing bacterium, for nitrogen fixation.[20] This work can be potentially extended to the development of supernitrogen fixing bacteria that can reduce the use of fertilizers. Indeed, synthetic biology has great potential; one day in the near future, we might hear something like this in the news:

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"Dr. Creatorgani, CEO of a synthetic biology company *Biocreaterix*, announced that the company has created a super gasoline-producing bacterium directly from carbon dioxide (Figure 1). It took them six months from designing the genome sequence on a computer to actually creating an artificial bacterium. They computer-designed the genome sequence by combining parts of the *E. coli* genome sequence, carbon dioxide fixing pathways of plants and newly designed hydrocarbon biosynthetic pathway genes. After initial cultivation studies, they optimized the gene expression levels at the whole metabolic and gene regulatory levels. This resulted in the production of a hydrocarbon mixture similar to gasoline with an impressively high volumetric productivity of 20 g/L/h by continuous fermentation. This created bacterium was named as '*Gasolinomonas efficientproduciens*'."

AUTHOR INFORMATION

Corresponding Author

*Tel: 82-33-343-1115. E-mail: jouna@minjok.hs.kr.

Author Contributions

G.N.L. and J.N. wrote the commentary together and read and approved the final version of the commentary.

Notes

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