Synthetic microbes -An overview of these multipurpose microbial tools

Reema Subramanian
PhD candidate
Supervisor: Professor Margaret Ip
Joint Graduate Seminar
Department of Microbiology
The Chinese University of Hong Kong
1st December 2016

Outline

1. Synthetic microbe - Concept and Science
2. Creating a synthetic microbe - What are the resources that we have and can use?
3. Application of synthetic microorganisms- Successful developments and future implications

Synthetic microbe

Concept and Science

Synthetic microbes

- Microbes are ubiquitous
- Relatively simple genomes- very well studied
- Relatively easy to manipulate
- Can be engineered to perform specific tasks
- Manipulate simplest life forms to solve problems on a global scale (Microbial machines)

Synthetic Microbes: ‘Organisms synthesized or engineered by human beings to perform specific tasks, that can grow, reproduce and develop without further human intervention’[1]

1 Modified from: Deplazes, Anna, and Markus Huppenbauer. “Synthetic Organisms and Living Machines: Positioning the Products of Synthetic Biology at the Borderline between Living and Non-Living Matter.” Systems and Synthetic Biology 3.1-4 (2009): 55–63. PMC. Web. 30 Nov. 2016.

Science behind synthetic microbes

- **Synthetic biology-** "Design and construction of novel biologically based parts, devices and systems or redesigning existing natural biological systems"
- Aim: Design an efficient biological system to achieve a specific desired output

- Two approaches to creating synthetic microbes:
  - Bottom-up approach: Creating life *de novo*
  - Top-down approach: Designing and inserting elements inside cells to perform specific tasks

Roberts, M. A. J. et al. "Synthetic Biology: Biology by Design." Microbiology159.Pt 7 (2013): 1219–1220. PMC. Web. 30 Nov. 2016.

Bringing ‘engineering’ back in genetic engineering

- Decades of genetic ‘manipulation’ through conventional methods
- Very little ‘engineering’ (Not much ‘designing’ and ‘building’ involved)
- Problems with conventional molecular biology techniques
  - Dated
  - Slow and time consuming
  - Lack of standardisation

  - Design and create standardised components that can be assembled easily to create new reliable things

TechNyouvids. "Synthetic Biology Explained." *YouTube*. YouTube, 09 Aug. 2011. Web. 30 Nov. 2016.

‘Creation’ as opposed to ‘manipulation’

Synthetic biology

Major drivers towards the development of synthetic biology- **ability to synthesize DNA**

Creating a synthetic microbe

What are the resources that we have and can use?

Synthetic microbes - the vision

- Microbes are ubiquitous
- Relatively simple genomes- very well studied
- Relatively easy to manipulate
- Can be engineered to perform specific tasks
- Manipulate simplest life forms to solve problems on a global scale (Microbial machines)
- DNA- software or instructions for making a specific product (genes for a function/genome)
- Living cell- 'Micro factory'
- Synthetic Microbes: 'Organisms synthesized or engineered by human beings, that can grow, reproduce and develop without further human intervention'1

1 Modified from: Deplazes, Anna, and Markus Huppenbauer. "Synthetic Organisms and Living Machines: Positioning the Products of Synthetic Biology at the Borderline between Living and Non-Living Matter." Systems and Synthetic Biology 3.1-4 (2009): 55–63. PMC. Web. 30 Nov. 2016.

Biobricks

- “Standard DNA sequences with defined structure and function and share a common interface”[1]
- Overcome lack of standardisation of traditional molecular biology methods
- Open source- used to assemble complex biological circuits from simple building blocks
- Introduced in 2003 (MIT)
- Biobricks foundation: Non profit organisation. Focused on further standardising and promoting these biological parts
  - Easily accessed and used by students!

1 ”SynBio Standards -BioBrick" (PDF). Retrieved 29 November 2016

Shetty, Reshma P, Drew Endy, and Thomas F Knight. “Engineering BioBrick Vectors from BioBrick Parts.” *Journal of Biological Engineering* 2 (2008): 5. *PMC*. Web. 30 Nov. 2016.

Biobricks

Parts

Building blocks. Encode basic biological functions

E.g.: Coding sequences, promoters, terminators etc.

Devices

Collection of Parts for a defined function.

E.g: Riboregulator producing a fluorescent protein in response to an environmental trigger

System

Combination of devices to perform a higher level task

E.g: Multiple fluorescent signals in response to multiple environmental triggers working in consortium

SynBio Standards -BioBrick" (PDF). Retrieved 29 November 2016

Synthetic microbes - the vision

- Microbes are ubiquitous
- Relatively simple genomes- very well studied
- Relatively easy to manipulate
- Can be engineered to perform specific tasks
- Manipulate simplest life forms to solve problems on a global scale (Microbial machines)
- DNA- software or instructions
- Living cell- 'Micro factory'- platform to run the 'software'

Synthetic Microbes: 'Organisms synthesized or engineered by human beings, that can grow, reproduce and develop without further human intervention'1

1 Modified from: Deplazes, Anna, and Markus Huppenbauer. "Synthetic Organisms and Living Machines: Positioning the Products of Synthetic Biology at the Borderline between Living and Non-Living Matter." Systems and Synthetic Biology 3.1-4 (2009): 55–63. PMC. Web. 30 Nov. 2016.

The ‘Minimal cell’

RESEARCH ARTICLE SUMMARY

SYNTHETIC BIOLOGY

Design and synthesis of a minimal bacterial genome

Clyde A. Hutchison III,*† Ray-Yuan Chuang,† Vladimir N. Noskov, Nacyra Assad-Garcia, Thomas J. Deerinck, Mark H. Ellisman, John Gill, Krishna Kannan, Bogumil J. Karas, Li Ma, James F. Pelletier, Zhi-Qing Qi, R. Alexander Richter, Elizabeth A. Strychalski, Lijie Sun, Yo Suzuki, Billyana Tsvetanova, Kim S. Wise, Hamilton O. Smith, John I. Glass, Chuck Merryman, Daniel G. Gibson, J. Craig Venter*

JCVI-syn3.0

- J. Craig Venter Institute (JCVI) and Synthetic Genomics, Inc. (SGI)
- Modified *Mycoplasma mycoides* genome
- Deletion of genes not essential for growth under laboratory conditions
- Genome **581 kbp, 473 genes-** smaller than any autonomously replicating cell in nature
- Whole genome design, Chemical synthesis and genome transplantation into *M. capricolum* recipient

Hutchison, C. A.,3rd, et al. "Design and Synthesis of a Minimal Bacterial Genome." *Science (New York, N.Y.)* 351.6280 (2016): aad6253. Print. 30 Nov 2016

The ‘Minimal cell’ approach for a more ideal chassis

Hutchison, C. A.,3rd, et al. "Design and Synthesis of a Minimal Bacterial Genome." *Science (New York, N.Y.)* 351.6280 (2016): aad6253. Print. 30 Nov 2016

https://en.wikipedia.org/wiki/BioBrick#/media/File:Standard_assembly_10.jpg(Modified)

Application of synthetic microorganisms

Successful developments and future implications

‘E.chromi’
(University of Cambridge)

- *E.coli* with a sensitivity tuner (biosensors) and colour generator (Biobricks)
- Turn into a certain colour when environmental toxin of a certain concentration is detected
- Potential application in diagnostics- Digestive tract disorders

http://2009.igem.org/Team:Cambridge/Project/Amplification

Synthetic yeast – micro factories for an antimalarial production

- **Artemisinin:** Antimalarial drug produced by the plant *Artemisia annua*.
  - Global supply exclusively by farmers that cultivate the plant- success highly variable
  - Production occurs naturally in this plant using sugars generated by photosynthesis
- **Professor Jay Keasling** (UC Berkeley)- cheap and reliable alternative to agricultural production
- Using engineered yeast for production of the antimalarial precursor at an industrial scale- > 25 g per Litre of artemisinic acid

Paddon, Chris,J., and Jay Keasling D. "Semi-Synthetic Artemisinin: A Model for the use of Synthetic Biology in Pharmaceutical Development." Print 30 Nov. 2016.

Production of biofuels

- Algae- natural production of high biomass
- Accumulate energy dense oils which can be used as fuels
- **Sythetic algae-** optimization of fuel production pathways
- Replace fossil fuels in the future?

Gimpel, J. A., et al. "Advances in Microalgae Engineering and Synthetic Biology Applications for Biofuel Production." *Current opinion in chemical biology* 17.3 (2013): 489-95. Print 30 Nov 2016

"The Algae Project." *Storybank* Web. 30 Nov. 2016.

Other applications

- Potential to revolutionise the manufacturing of ‘everyday products’

  Detergents, cosmetics, food etc.
  - Terpenes- Work towards producing synthetic yeast for industrial levels
  - Bioacrylic – **OPXBIO.** Application in detergents, paints etc.

- Living therapies/vaccines
  - **EnBiotix**- EP001 phage- target bacteria to produce biofilm destroying enzymes
  - **Prokarium-** ‘tamed’ Salmonella, enters immune cells, but does not cause disease

- Environmental
  - Biosensors for pollution detection?

Tippmann, S., et al. "Production of Farnesene and Santalene by Saccharomyces Cerevisiae using Fed-Batch Cultivations with RQ-Controlled Feed." *Biotechnology and bioengineering* 113.1 (2016): 72-81. Print 30 Nov 2016

"Synthetic Biology's Clinical Applications." *Science | AAAS* 14 Sept. 2016. Web. 30 Nov. 2016.

Why have we not moved forward with synthetic microbes?

- **Currently time consuming and high costs**
  - Decades of research required
  - Although continuously falling, costs of synthesizing DNA still high
- **Technology is not ready**
  - Artimisinin (Been around for over a decade, but never made it to market)
- **Bioethical factors (!!)**
  - Difference of opinions amongst the general public
  - Creation of "Frankenstein's monsters' (unnatural beings)
- **Potential risks**
  - Accidental release into the environment –
    - cannot predict how it would adapt and affect the biosphere
    - 'Unnatural organisms'
  - Increased threat of bioterrorism- Deliberate misuse

Costs of DNA synthesis rapidly declining, strict risk management protocols and measures and the inevitable increase in demand of their products, utilisation of synthetic microbes may be the way forward in the future

"Do the Benefits of Synthetic Biology Outweigh the Risks? | Genetic Literacy Project." *Genetic Literacy Project*., 19 Nov. 2013. Web. 30 Nov. 2016.

Summary

- Synthetic Microbes are any non-biological organisms capable of self replication
- Microbial factories can be designed for optimal function using synthetic biology
- Availability of resources like “BioBricks” will largely facilitate development of synthetic microorganisms
- Development of “minimal cells” for further optimisation of production
- Despite the risks, these multipurpose microbial tools hold great promise in the future, potential to replace products derived from scarce resources

Thank you!