

Synthetic biology kit:

The purpose of this kit is to introduce you to the world of synthetic biology. You will have an opportunity to manipulate bacteria and get an insight of their features.

What is synthetic biology about?

Synthetic biology is a relatively new field in biology and bioengineering. Its purpose is to create useful engineered biological systems or devices. But how does one “engineer” living organisms? There are fears and ethical controversies about such a subject. However the potential of this field is huge. Synthetic biology is not about cloning humans. It is about putting together genes present in some species to reach new useful functions, or about designing new DNA sequences to enhance or reduce genetic expression of some critical molecules. Synthetic biology not only gives the opportunity to create new biological devices, but it also offers a chance to understand cellular mechanisms better by “playing” with small DNA sequences and observing what is produced.

There are multiple examples of useful biological devices such as biological sensors (for medical diagnostic as well as for environmental pollutants), controlled drug delivery systems, environment “cleaner” and other more or less fancy applications...

Why work with bacteria?

Bacteria have a really bad reputation, but most of them are non-pathogenic strains, as for example the billion of bacteria that are living in symbiosis with us inside our gastro-intestinal track... The most used and best documented organism in synthetic biology is *Escherichia Coli* (*E. coli*), a bacteria naturally present in our gut. Some strains are harmful while others are not. In this kit, we provide you with a strain of non-pathogenic *E. coli*, the K12 strain.

If some bacteria are infectious while others are not, it is because they don't carry the same genetic information. Indeed, some pieces of DNA can be mutated, removed or added, leading to different phenotypes (what can be observed in the aspects or behaviours of the bacteria, what proteins the organism will produce or not).

What is iGEM?

The iGEM is a worldwide synthetic biology competition between teams from universities or engineering schools all around the world, trying to create new devices using their skills and imagination. The concept of Biobrick (a small portion of DNA that has a precise function and can be associated with others in order to create the desired device, like you can build something different with the same LEGO pieces) is central to this competition and to synthetic

biology in general. Each team can use Biobricks created by previous teams and has to design new Biobricks that could be useful for further iGEMers. This "open source" concept of the competition makes it a great platform for synthetic biology, especially for young motivated future researchers.

An iGEM high school competition has been created recently, so check whether you can create a team with your school if you are interested and motivated! You would be the first in Switzerland to take part in this adventure.

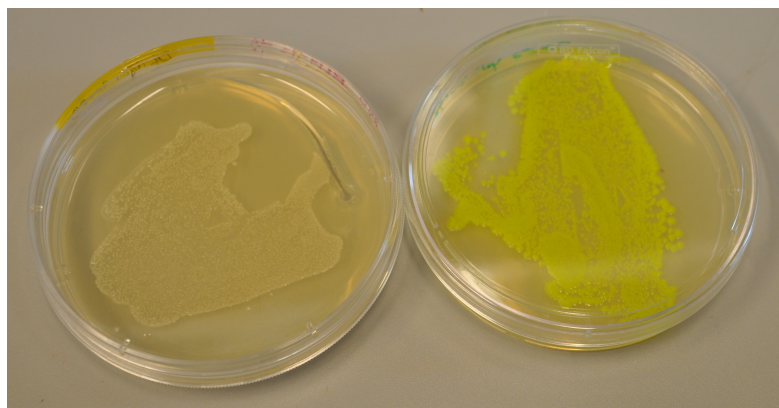
Here is the link to the iGEM website, where you can find informations about the contest, a Registry with all characterized parts (Biobricks) and links to the wikis of all previous and current projects: http://2013.igem.org/Main_Page.

(Taxi.Coli, EPFL team 2013, wiki: http://2013.igem.org/Team:EPF_Lausanne.)

The content and purpose of this kit:

In this kit, you are going to transform a bacterial strain with a Biobrick from the iGEM registry. EPFL team 2013 created this Biobrick, which is a plasmid carrying a resistance gene to an antibiotic (chloramphenicol) and expressing GFP, the green fluorescent protein, at a very high level.

You will start with a strain of non-pathogenic **competent** bacteria and a small sample containing a **plasmid** encoding for the Biobrick of interest. You will **transform** this plasmid into the bacteria. By plating some of your bacteria on a chloramphenicol containing Petri dish, you will select for the successfully transformed bacteria. Those will express GFP and hence appear green. The remaining bacteria will be plated on a Petri dish without antibiotic. On this non-selective plate, the rare transformed cells won't be seen: the non-transformed bacteria are much more numerous and you will therefore see only the "normal" white colonies they form.



Did it work? Well done, you made your first steps in the world of synthetic biology!

*You don't get the terms in bold? That's probably because you need a bit of theoretical background. This will be provided in the glossary section.

Except being fun, what is the interest of such a system?

Colorful bacteria are not, as such, useful. However, coupling the color-expressing gene to a sensing one can become very useful. You can create biosensors and detect some substances (toxins or pollutants) in a quite easy way. Of course, you can't just take bacteria in your hand, throw them on the floor and see whether a given pollutant is present or not. But by engineering further devices (e.g. semipermeable beads or more complex systems), you can prevent the bacteria from spreading and still keep their detecting capacity. And you can also modulate the effector part of the system, replacing the expression of the coloured protein by, for instance, the production of a useful molecule.

That's the magic part in synthetic biology: starting with some funny and fancy properties, you can modulate the genetic package of an organism to obtain useful and powerful devices! You can go as far as your imagination takes you, the only border you encounter is the one provided by nature...

Glossary

- Competent:** a competent cell is a cell that is able to incorporate a foreign **plasmid**.
- DNA:** deoxyribonucleic acid. Sequence of nucleic acids, called bases, that form a double-stranded helix and contain the genetic information of an individual. The whole DNA of an individual, called the genome, contains genes (only 2%), their associated **promoters** and also lots of other sequences which roles remain more or less understood by scientists (e.g. regulation, preservation,...).
- Plasmid:** a plasmid is a circular **DNA** fragment found in bacteria.
- Promoter:** **DNA** sequence generally present before or at the beginning of a gene that determines when in in what amount a gene is **transcribed**.
- RNA:** ribonucleic acid. Very similar to **DNA** except that there is one different base and that it stays in a simple non helicoidal strand.
- Transcription:** first major step in the expression of a gene in a protein, during which the gene **DNA** is copied in a **RNA** strand to allow further **translation**.
- To transform:** treat a bacterium in a way that makes it incorporate a given **plasmid**. Once the **plasmid** is inside the bacterium, it is **transcribed** and **translated** if it contains a **promoter** that is recognized by the bacterium.
- Translation:** second major step that brings from a gene to a protein using the **transcribed** copy of this gene.