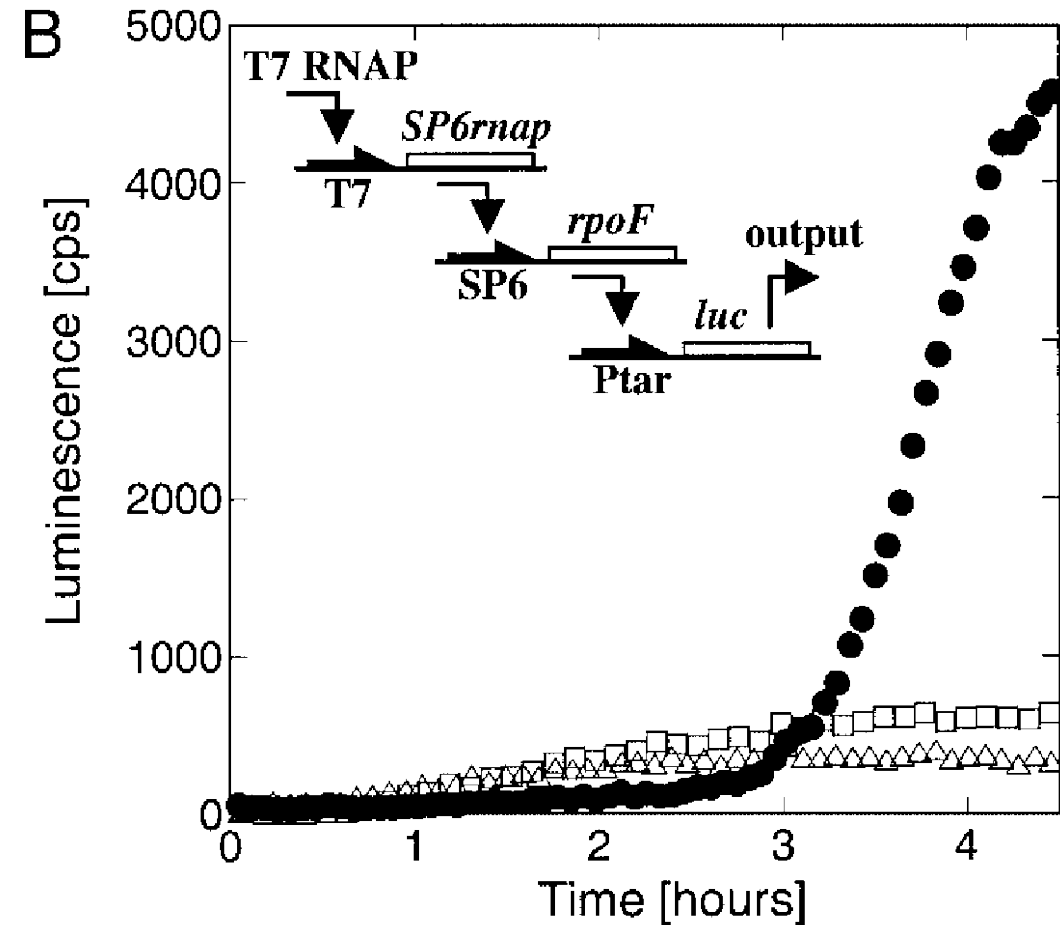
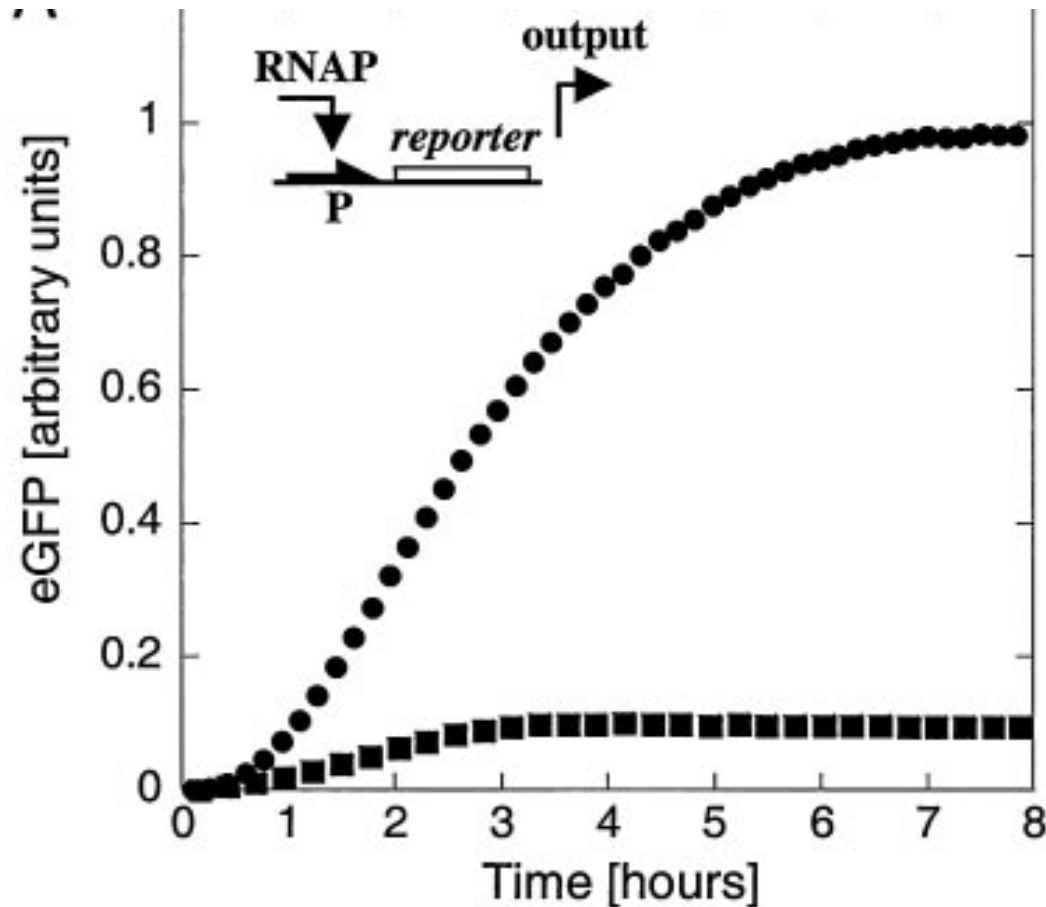


An Introduction and Overview of Cell-Free Synthetic Biology

Prof. Sebastian Maerkl

Cell-Free Synthetic Biology ?



Cell-free transcription – translation systems

Cell-based (lysate) system



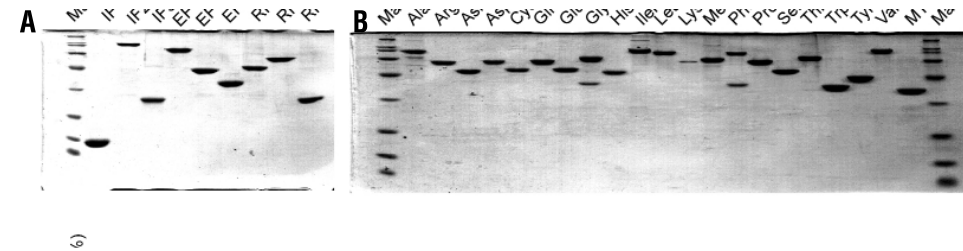
Pros:

- Highly active / high protein synthesis yields
- Cheap and easy to make
- Many different systems available

Cons:

- Undefined / complex system

PURE (recombinant) system



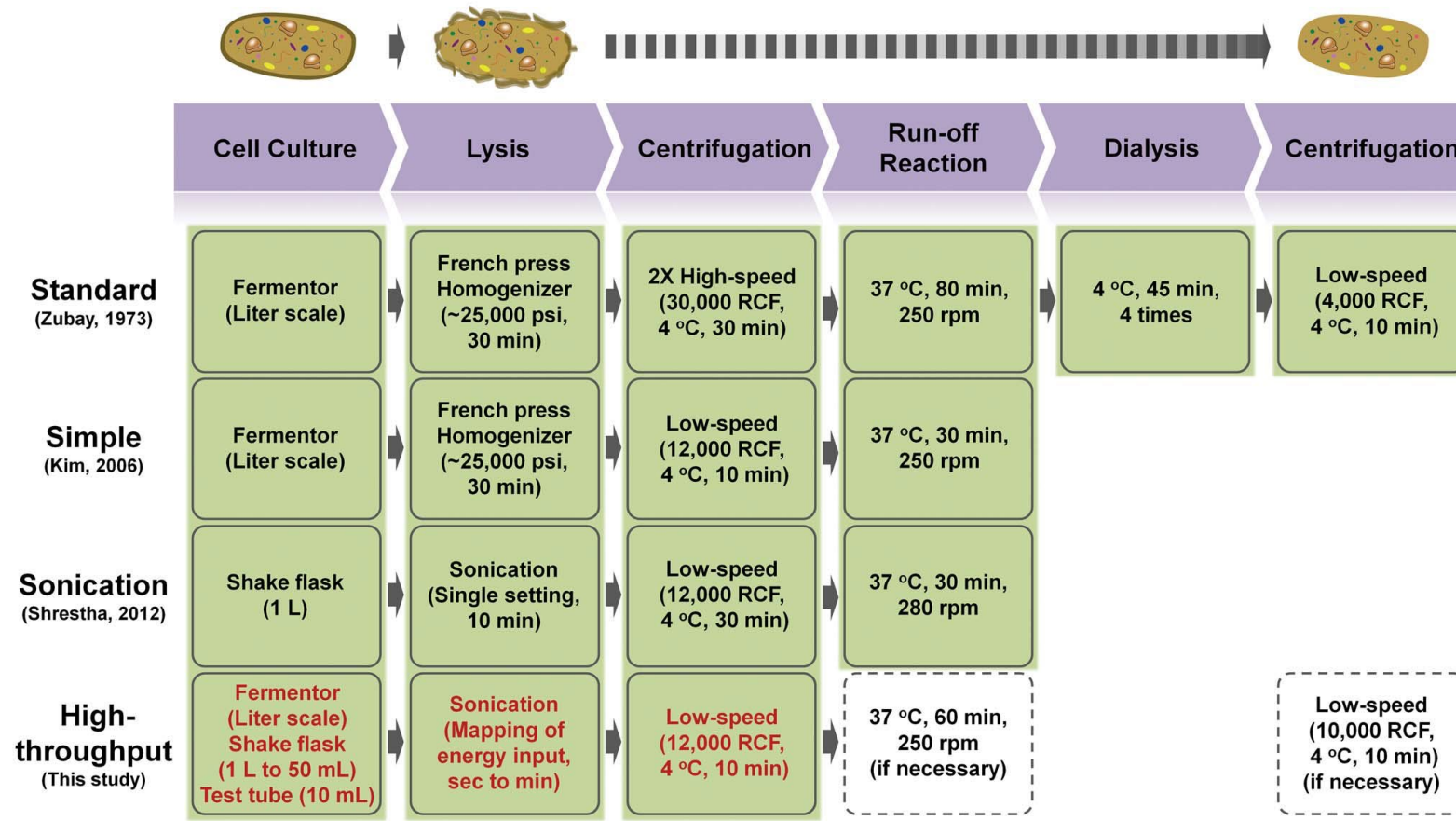
Pros:

- Completely defined system

Cons:

- Less active as lysate based systems
- Expensive and cumbersome to make
- “only” E. coli based system thus far

Lysate Based Systems



Lysate Flavors

Commercially available flavors:

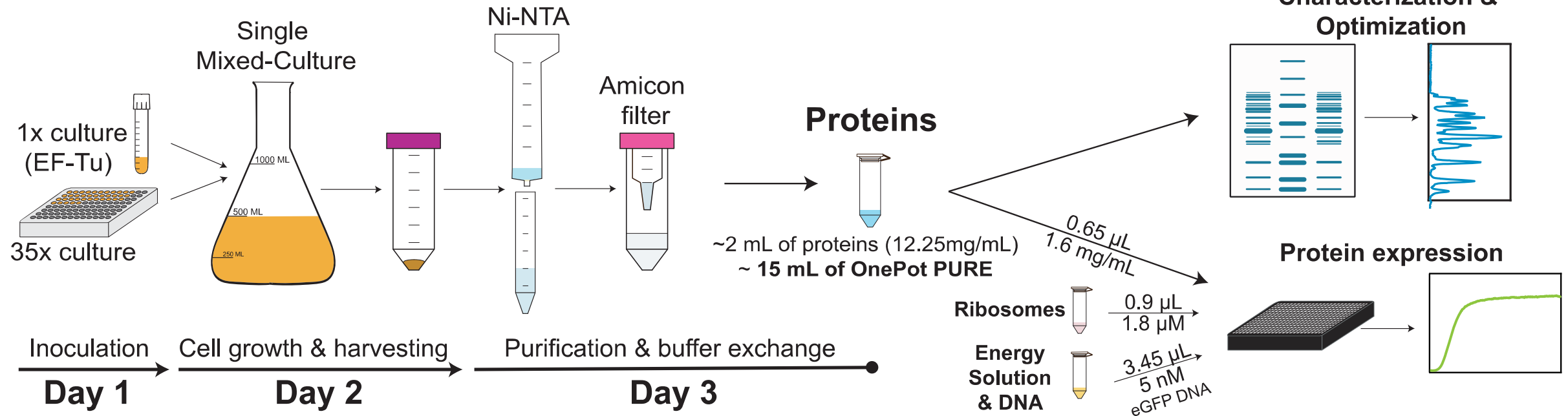
- E. coli
- Wheat germ
- Rabbit reticulocyte
- Mammalian

Additional flavors:

- V. natriegens
- B. subtilis
- B. megaterium
- E. fergusonii
- S. enterica
- K. oxytoca
- P. agglomerans
- P. putida
- C. glutamicum
- L. lactis

PURE System

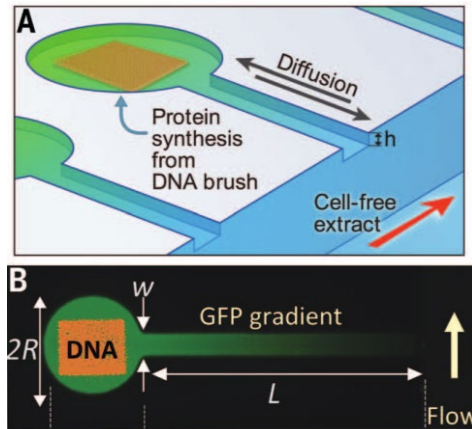
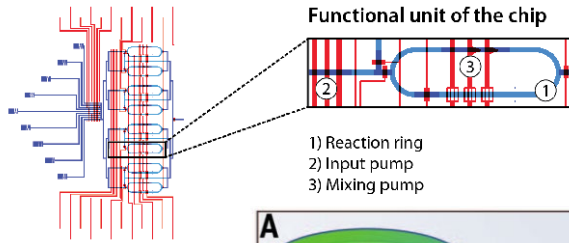
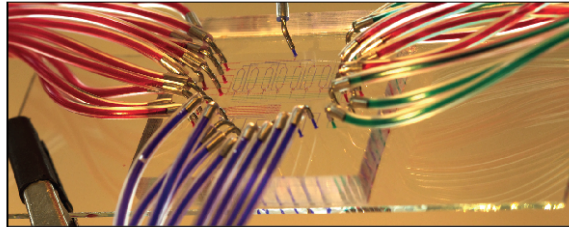
a OnePot Protein Purification



- New easy, cheap, and robust production method
- Individual protein components can be individually adjusted
- PURE system can be extended by addition of proteins / other components

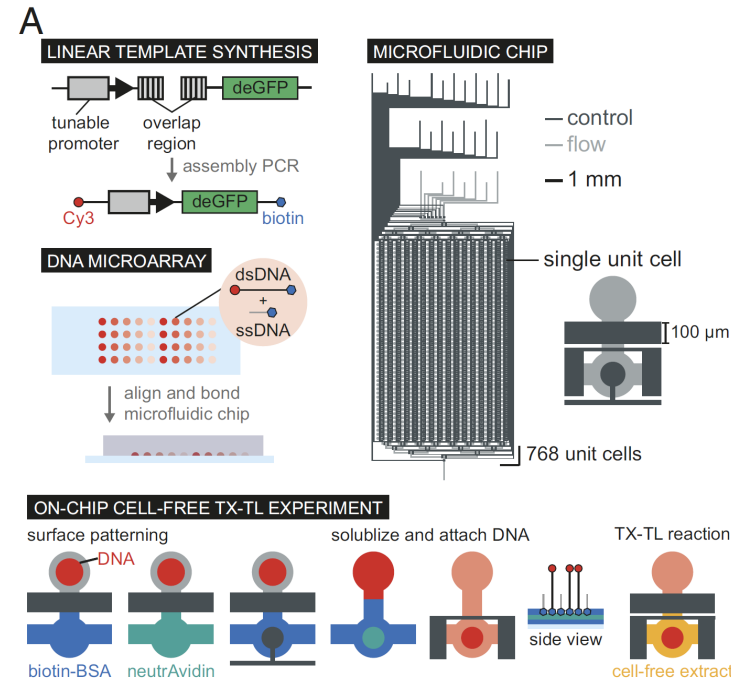
Cell-Free SynBio and Microfluidics

Chemostats



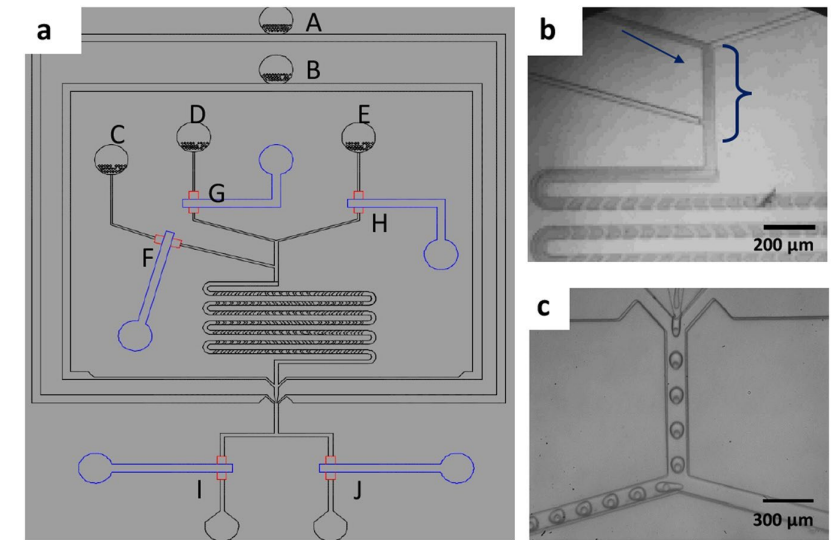
Niederholtmeyer et al. PNAS 2013.
Karzbrun et al. Science 2014.

High-throughput



Swank et al. PNAS 2019.

Droplets



Chang et al. SciRep 2018.

Is Cell-Free Synthetic Biology good for anything?

Basic science

Rapid Prototyping

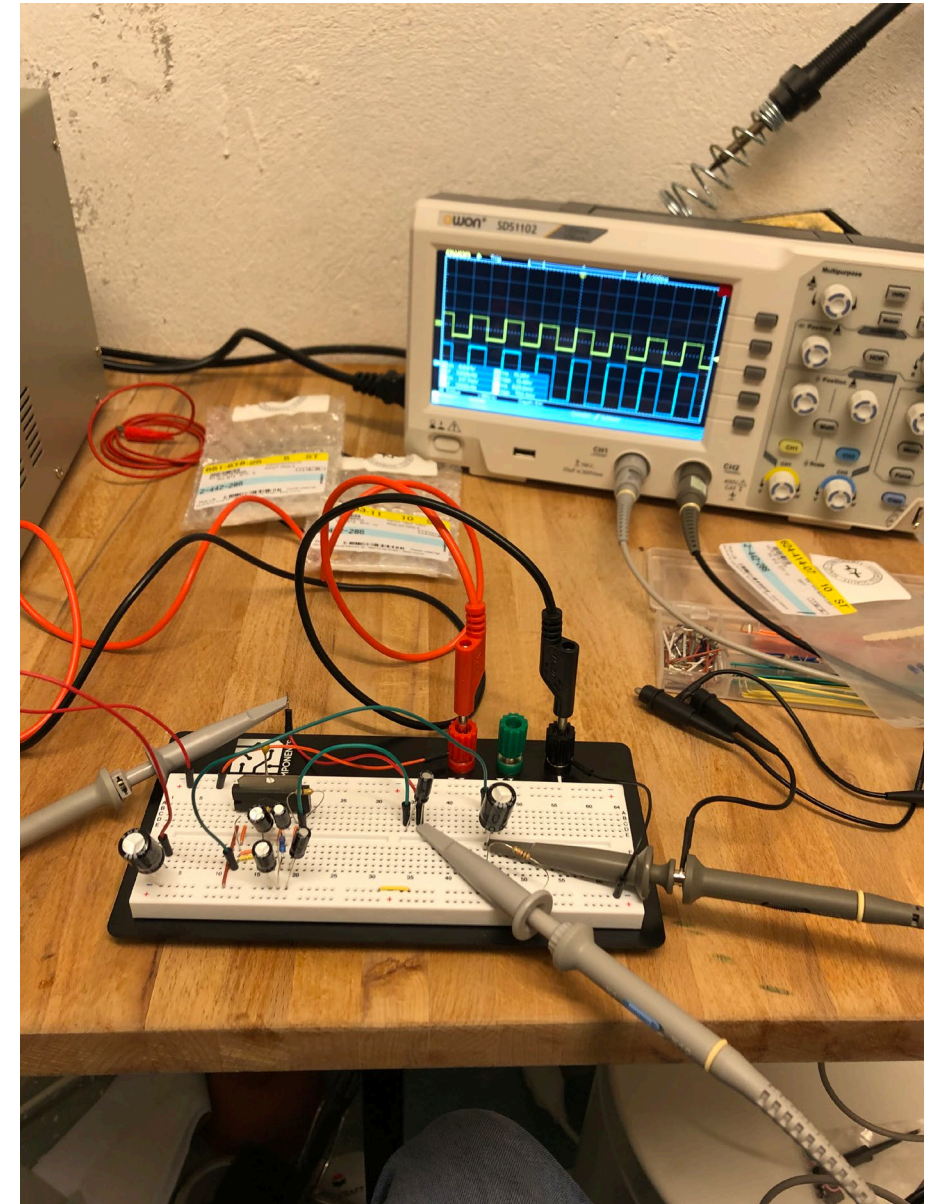
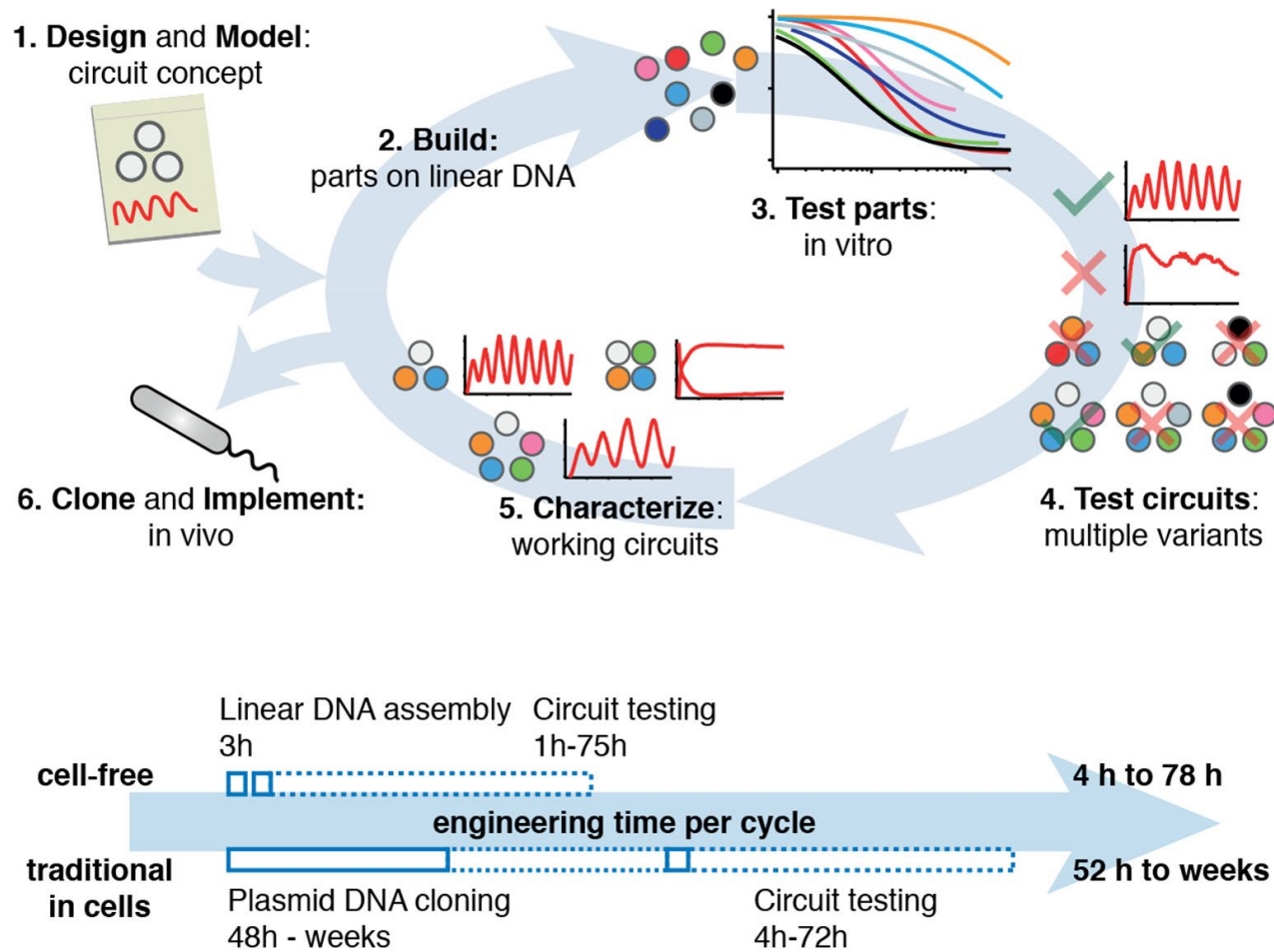
Metabolic Engineering

Sensing / Diagnostics / Therapeutics

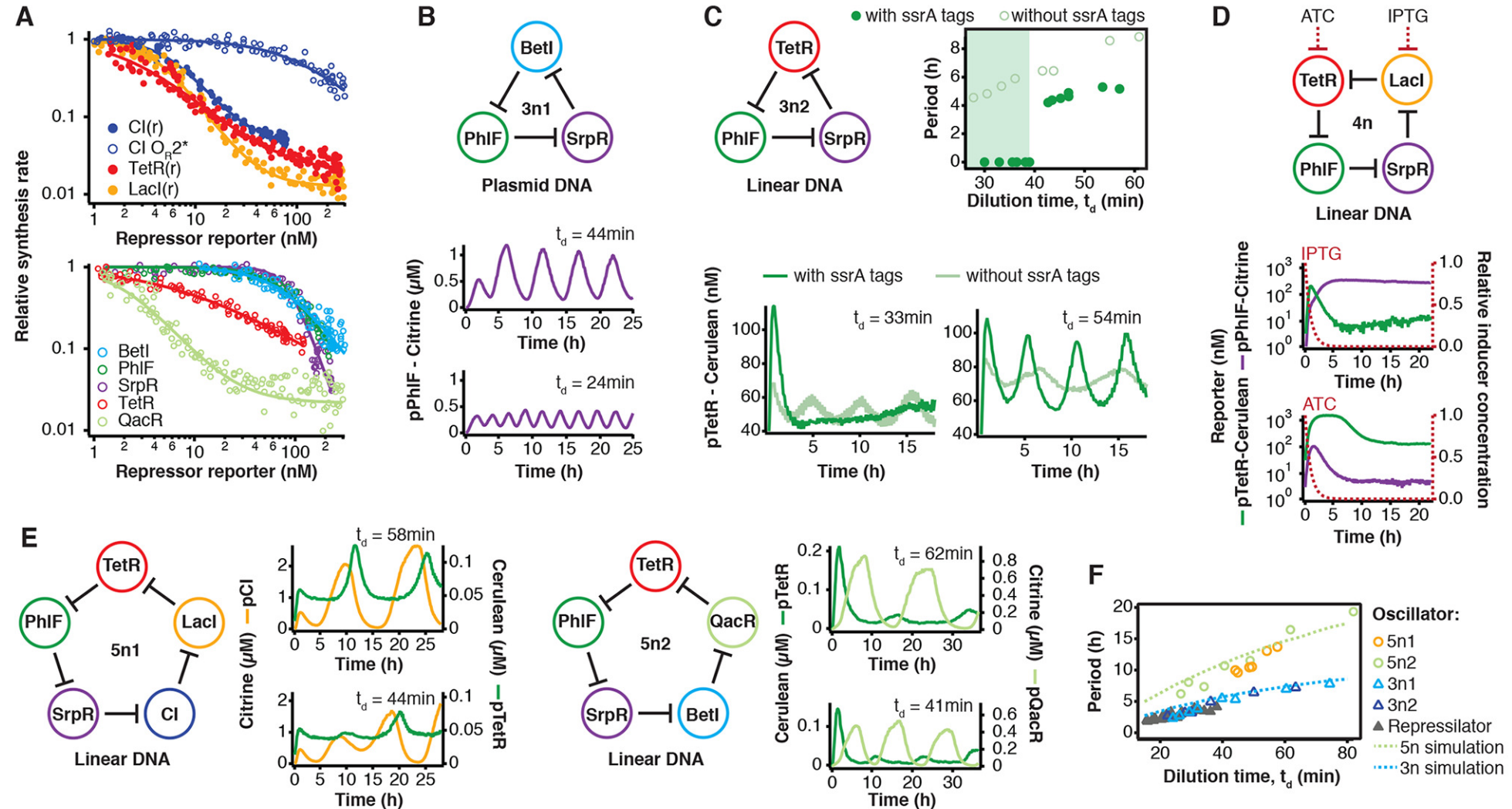
Education

Foundation for the development of an artificial cell

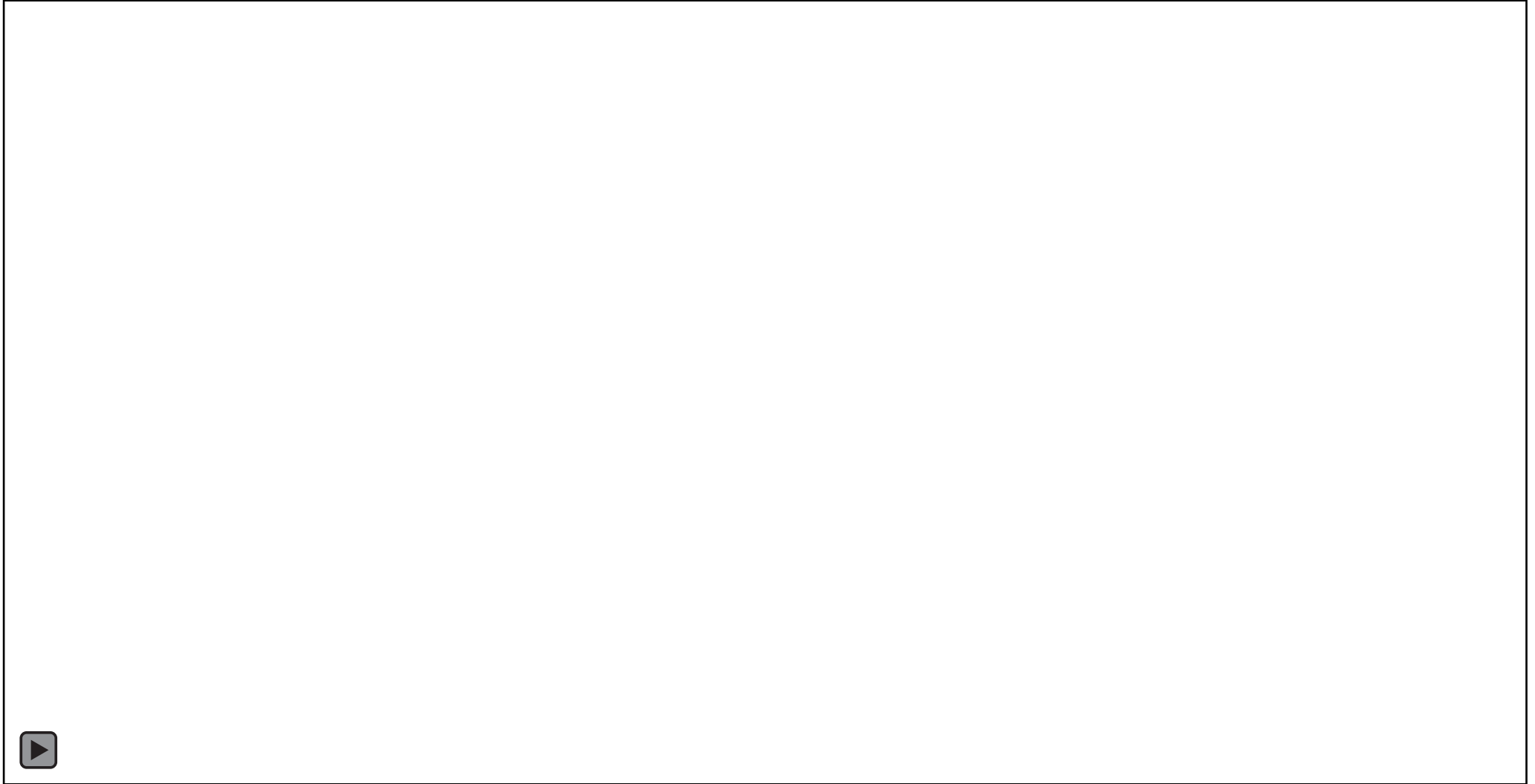
Rapid prototyping



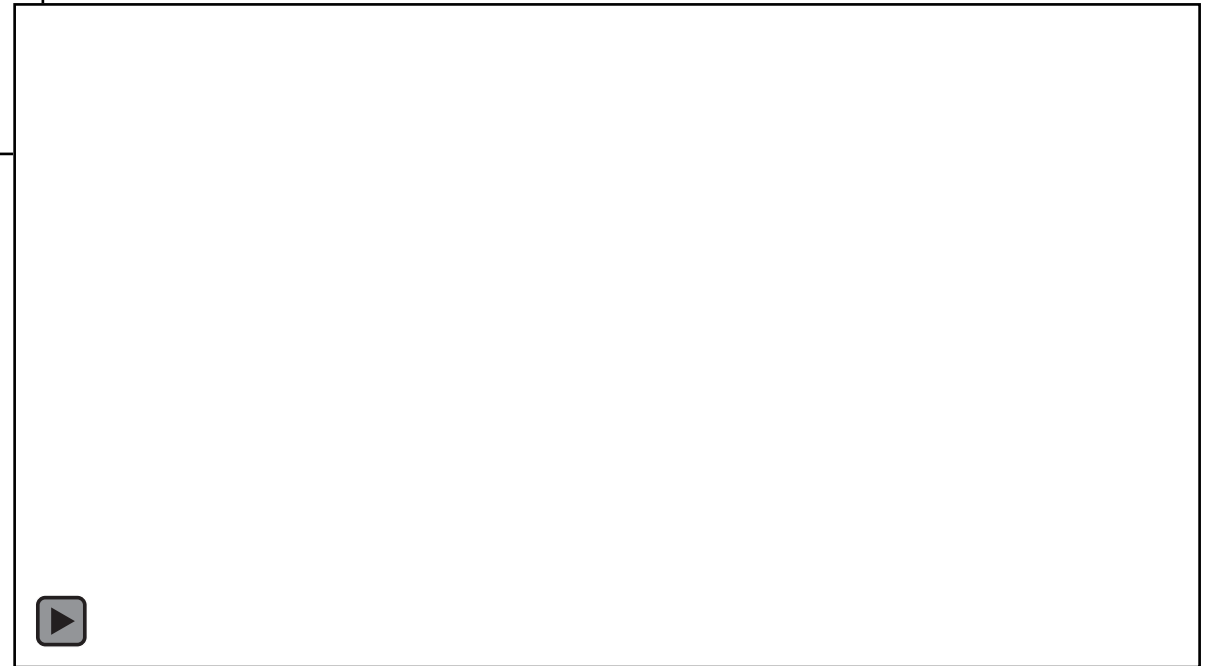
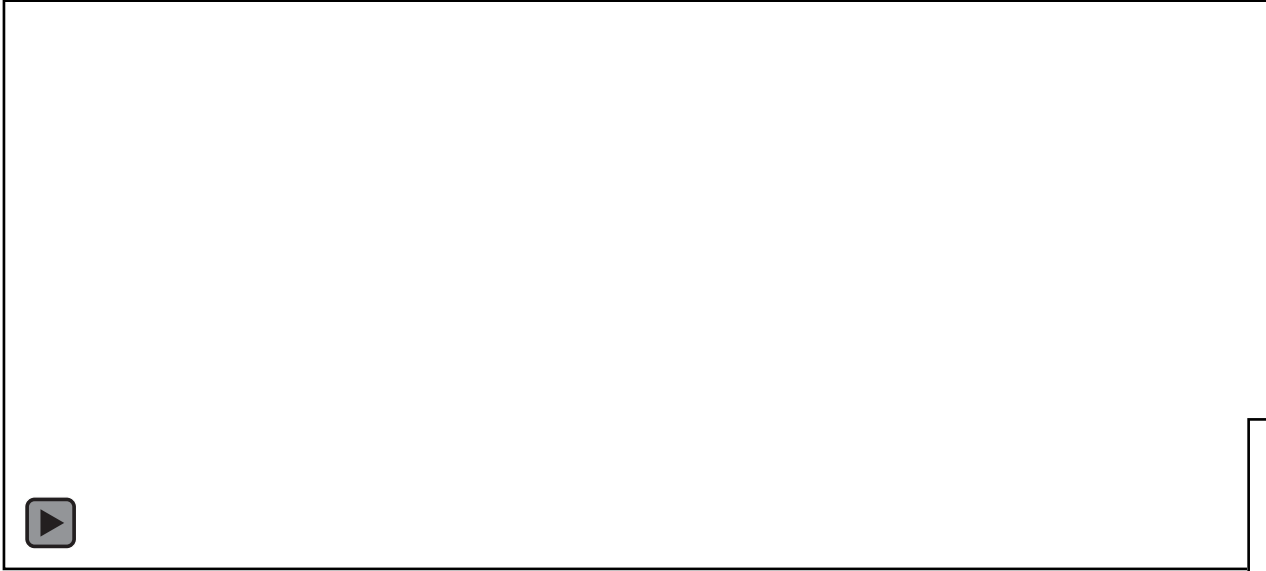
Rapid prototyping



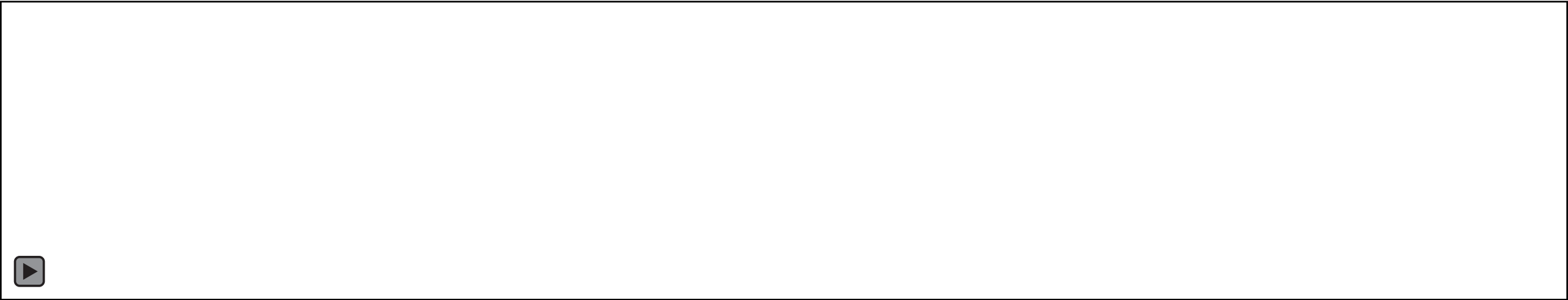
In vitro repressilator on a chemostat



In vivo 3n and 5n oscillators on a mothermachine



In vivo 3n oscillator on a cellasic device

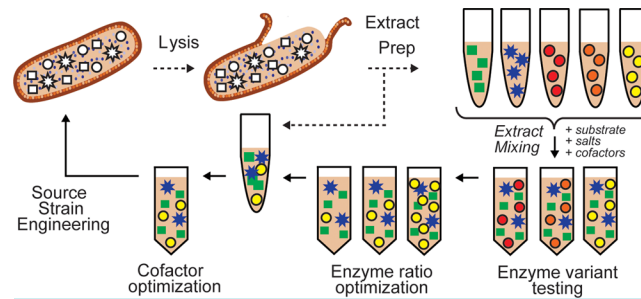


Metabolic Engineering

Non-ribosomal peptide synthesis

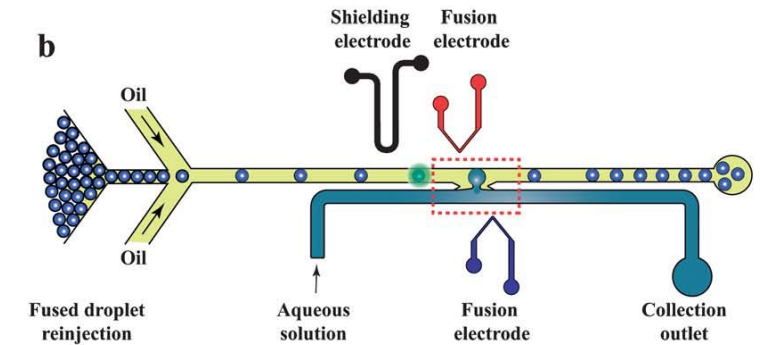
Goering et al. ACS SynBio 2017.

Pathway optimization



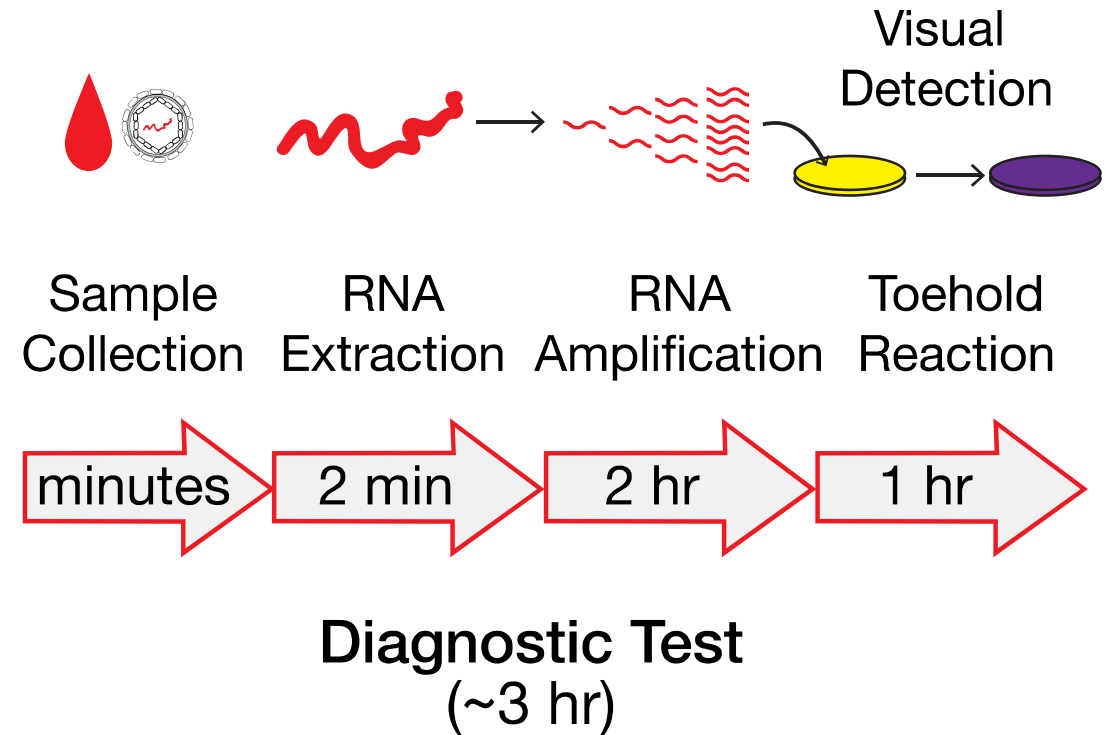
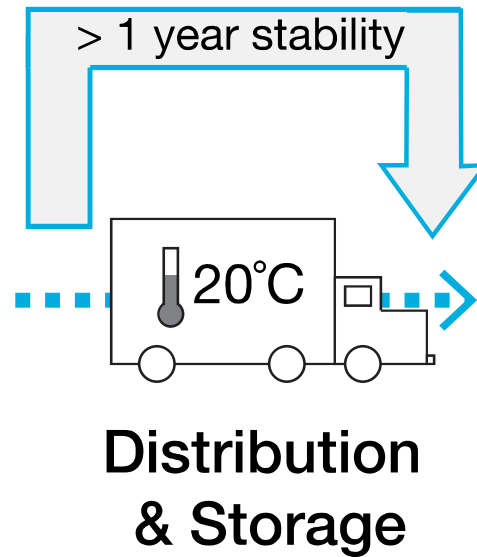
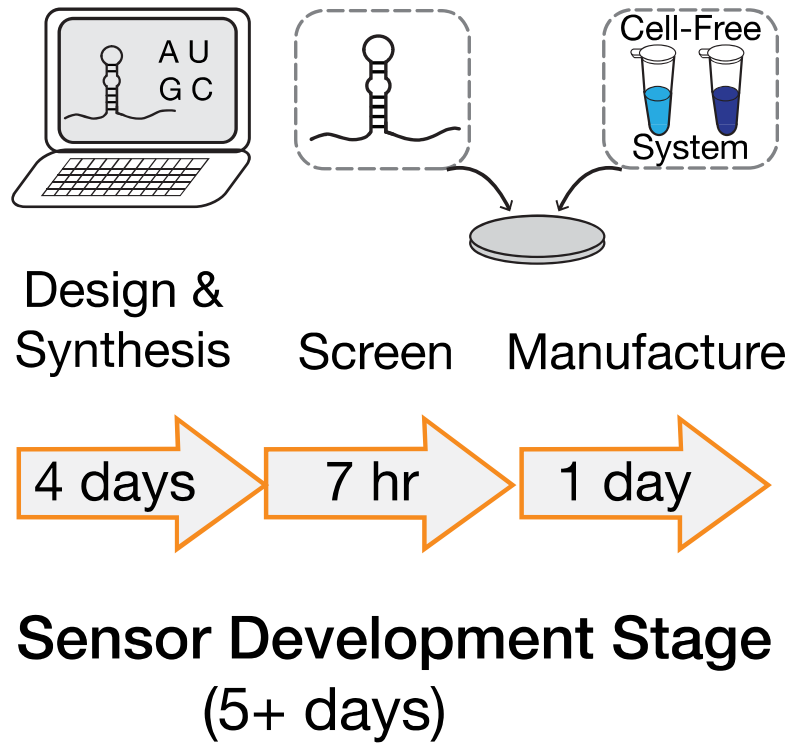
Dudley et al. ACS SynBio 2016.

Enzyme engineering / directed evolution

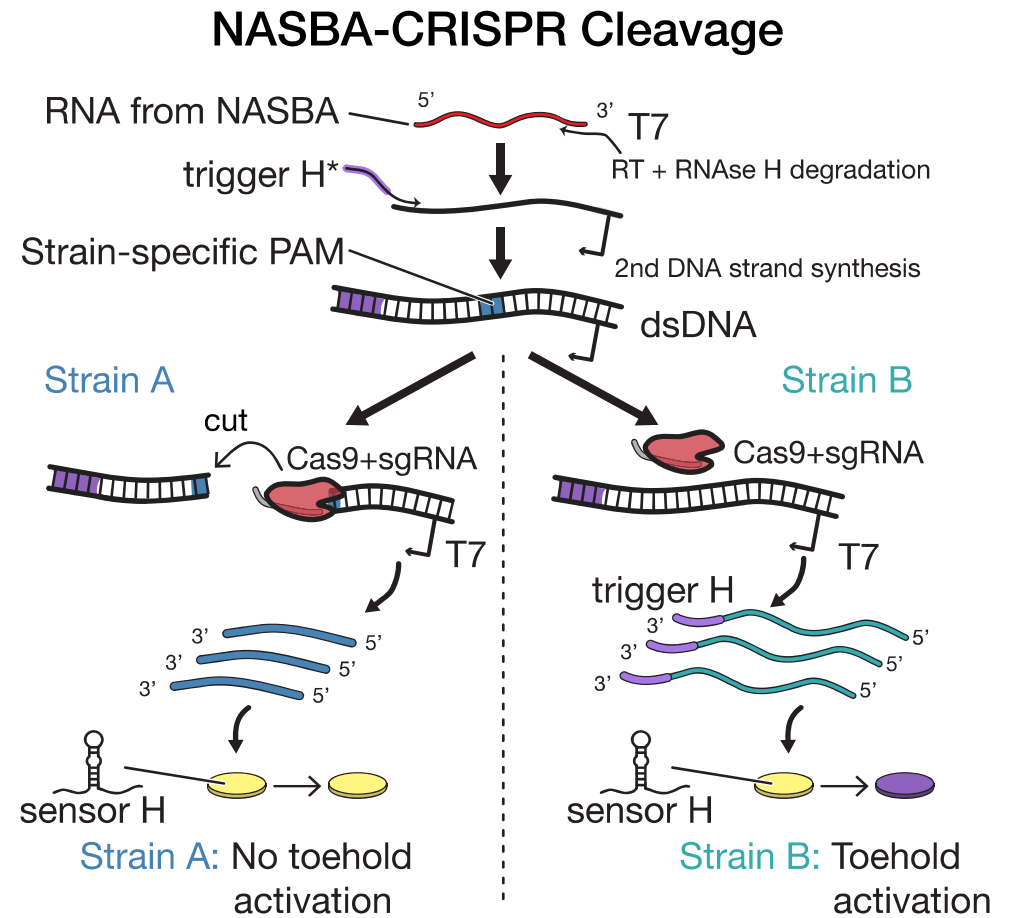
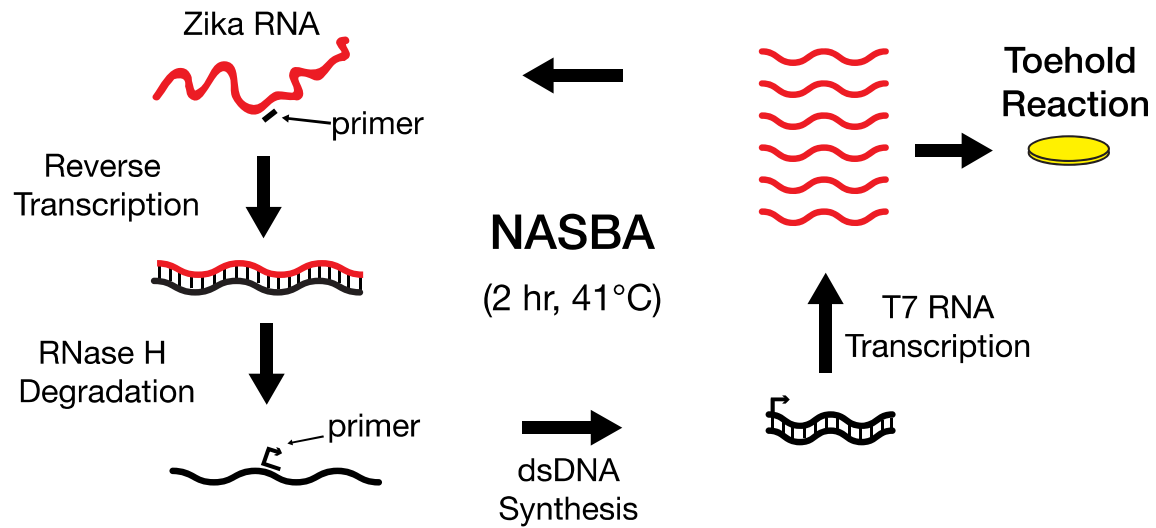


Fallah-Araghi et al. LOC 2012.

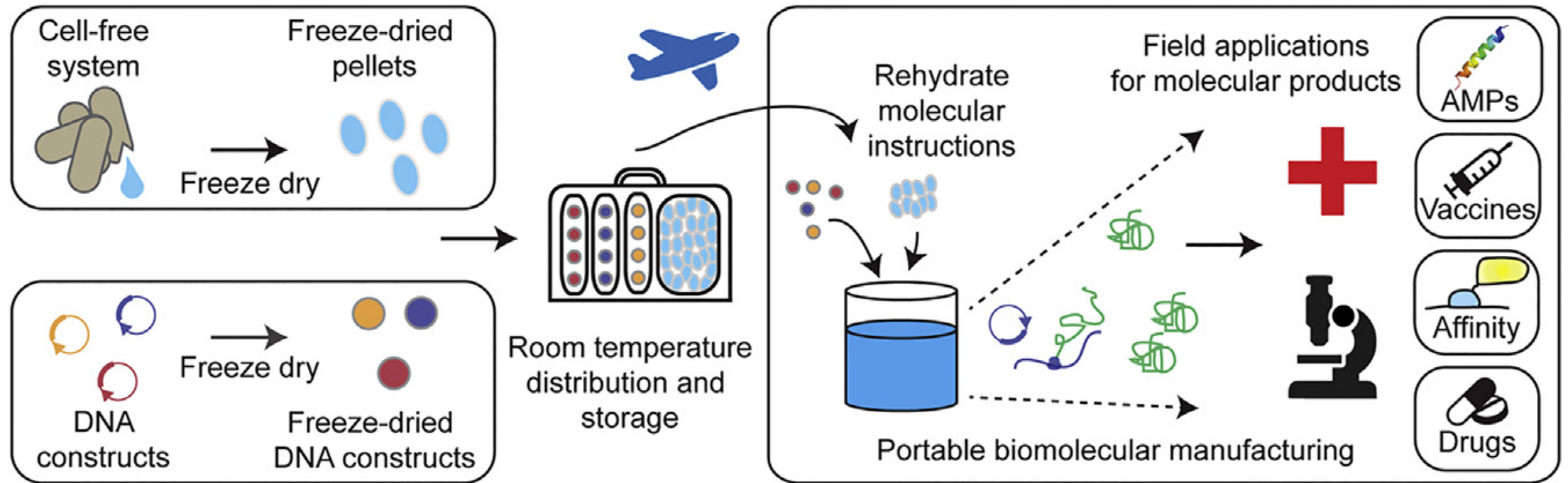
Diagnostics



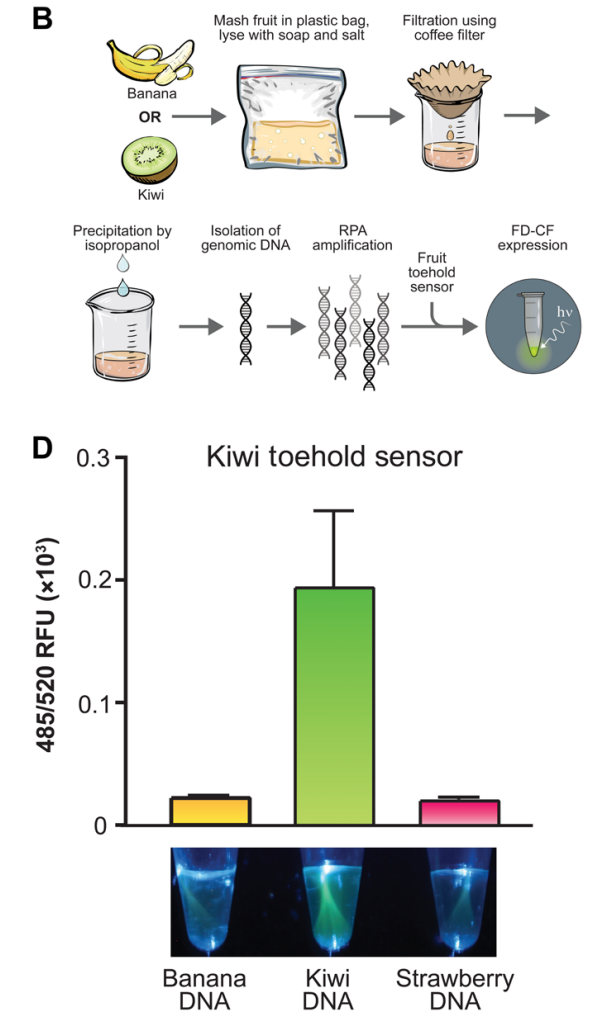
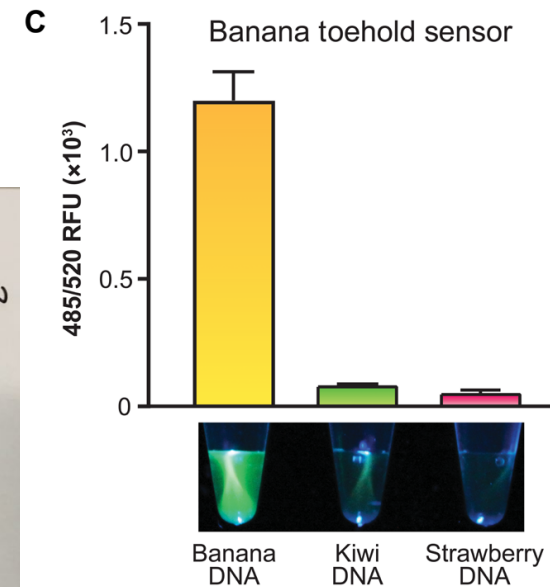
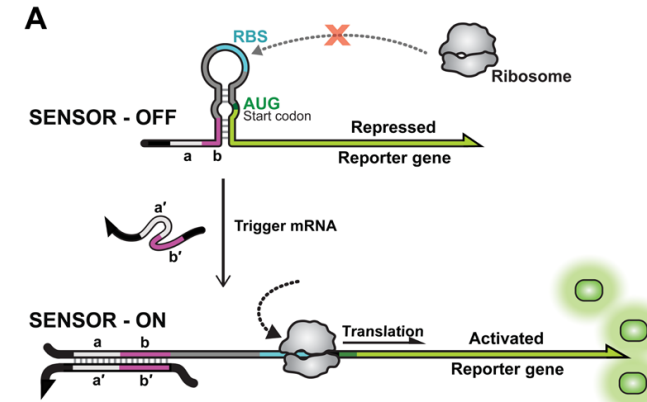
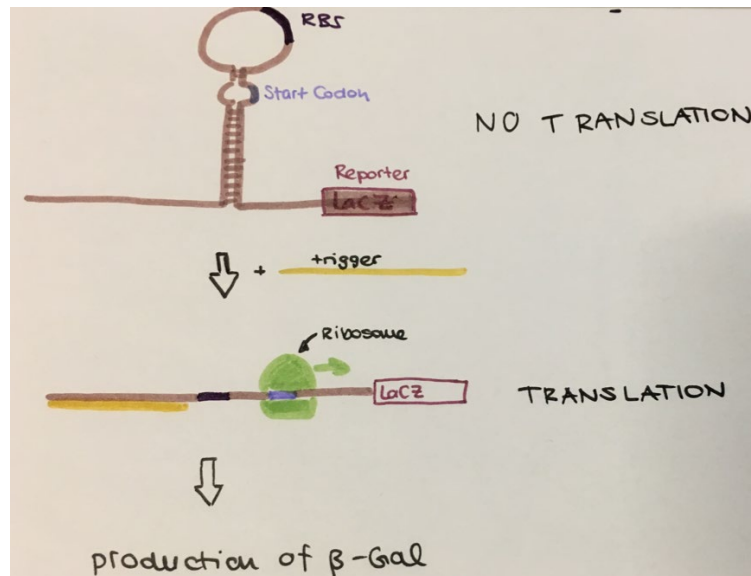
Diagnostics



Therapeutics / Biomolecular Manufacturing

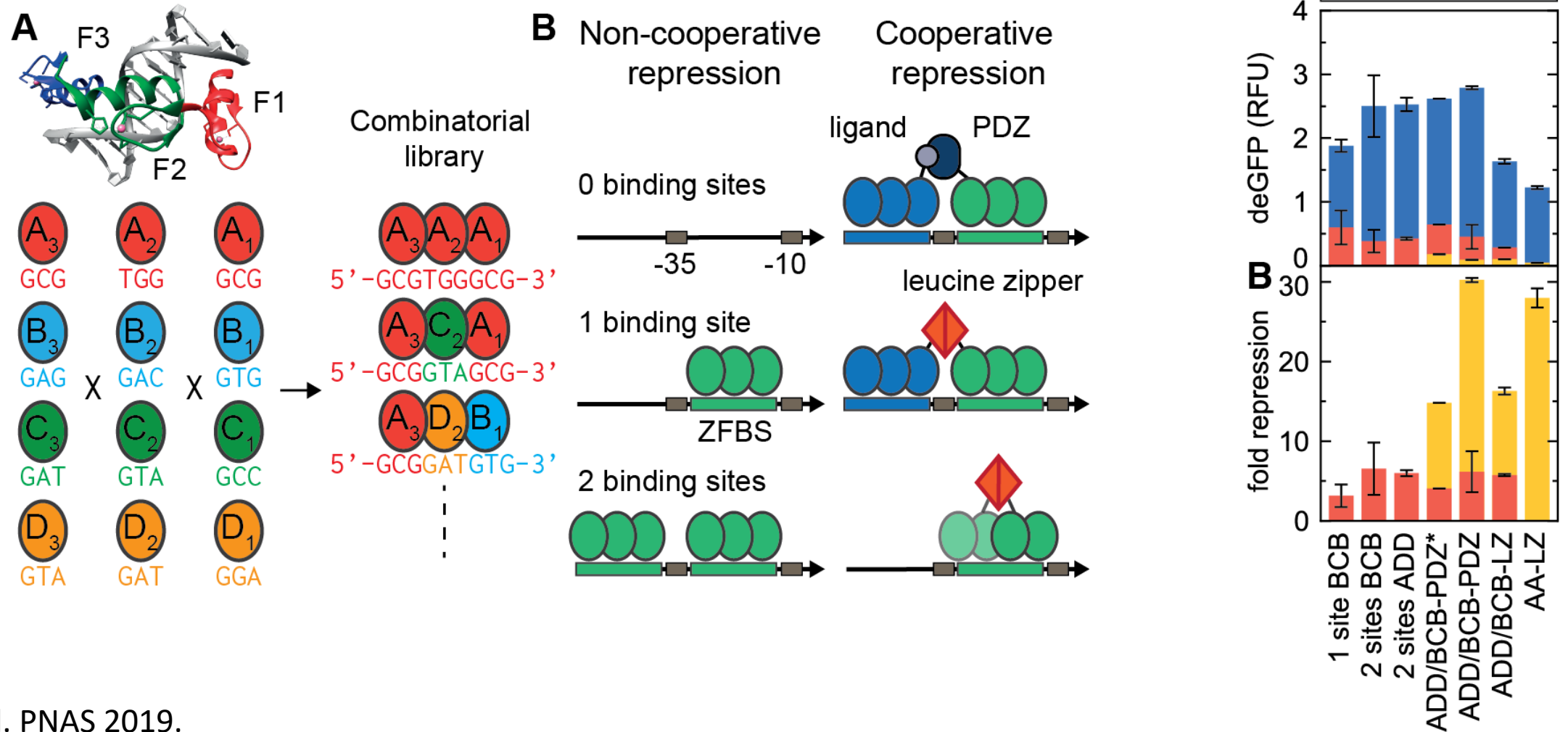


Education

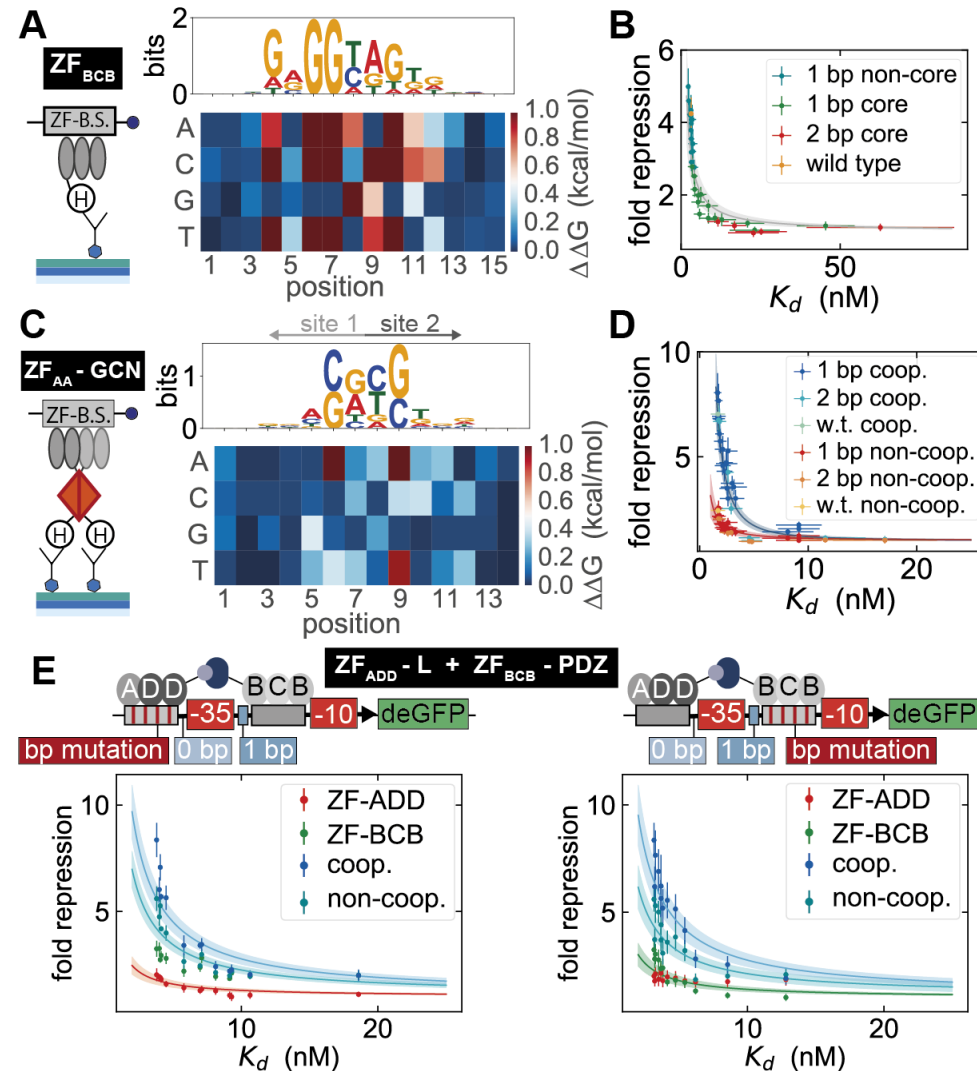


Stark et al. Science Advances 2018.
Huang et al Science Advances 2018.

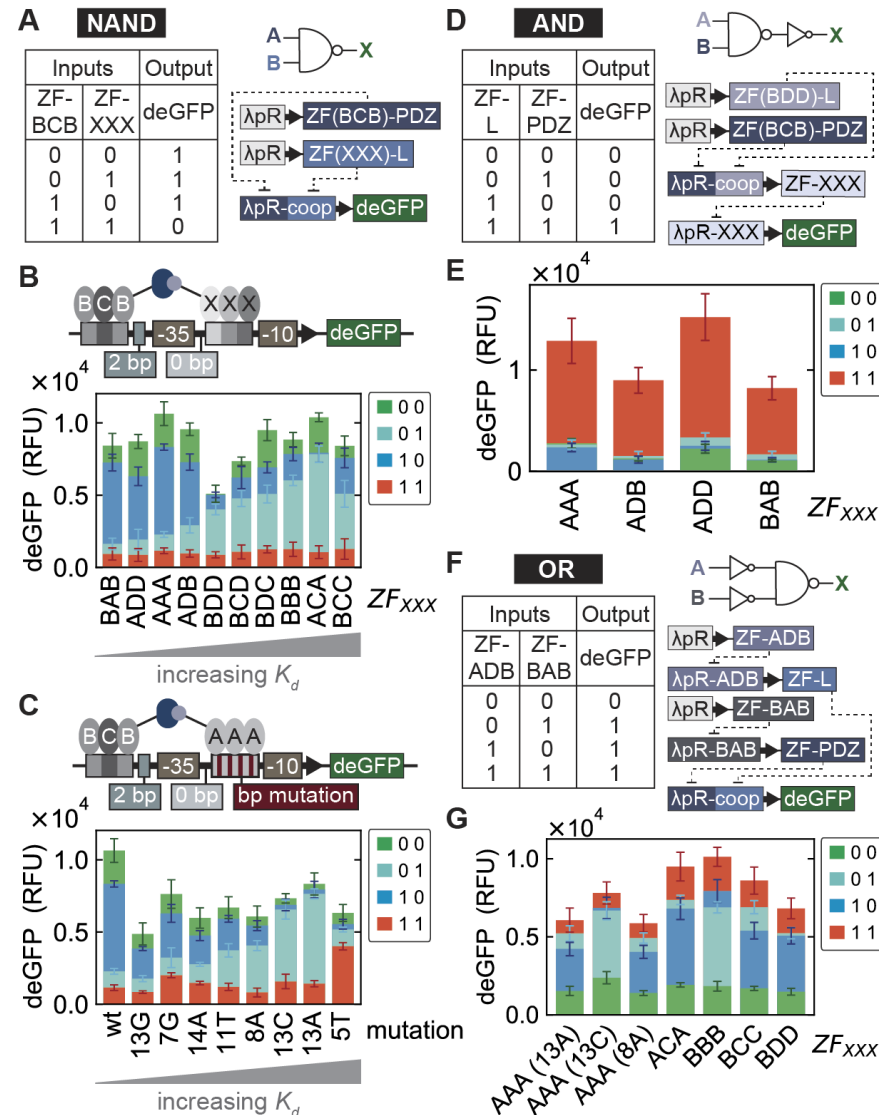
Bottom-up design, characterization, and implementation of gene regulatory networks.



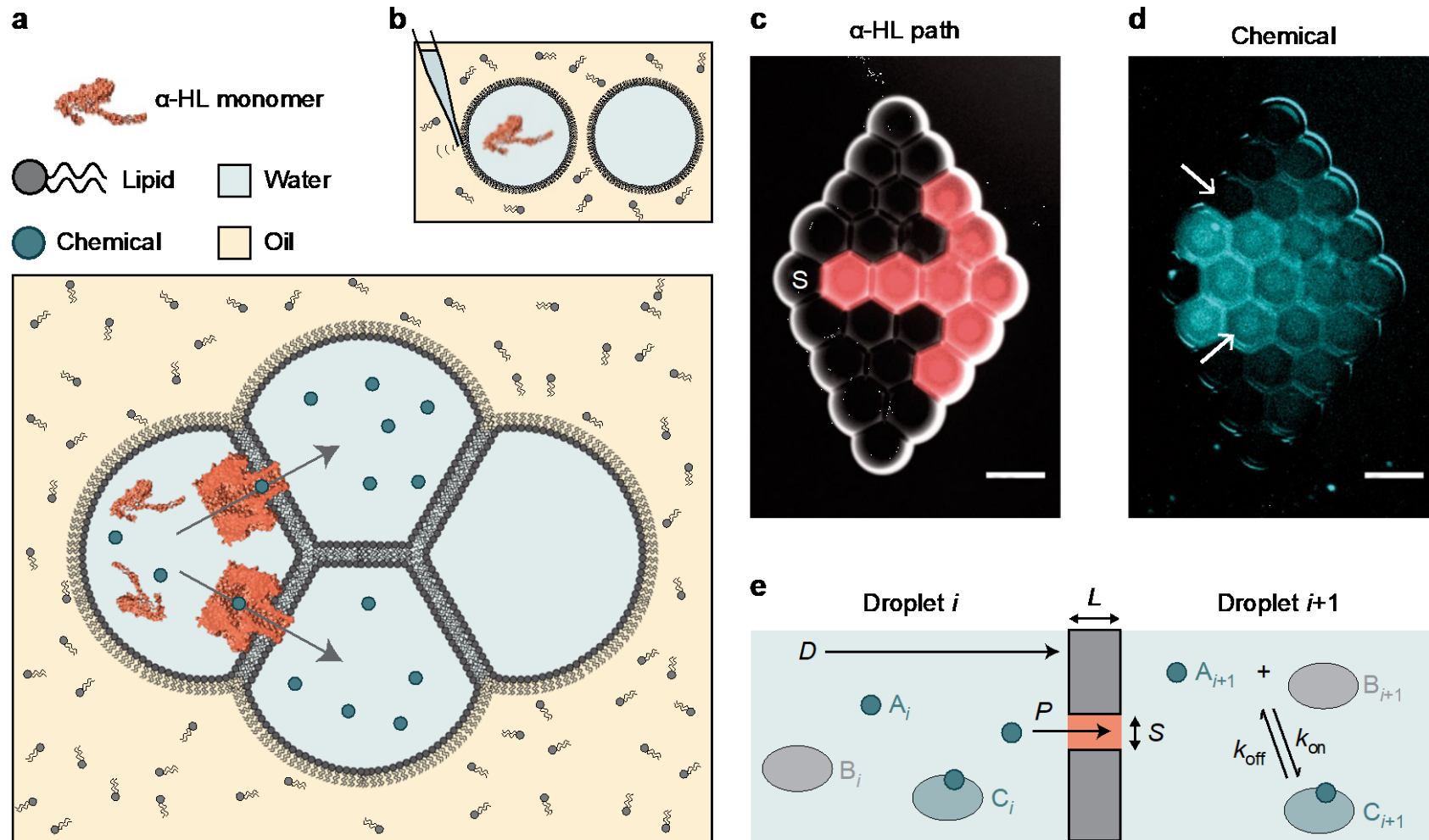
Bottom-up design, characterization and implementation of gene regulatory networks.



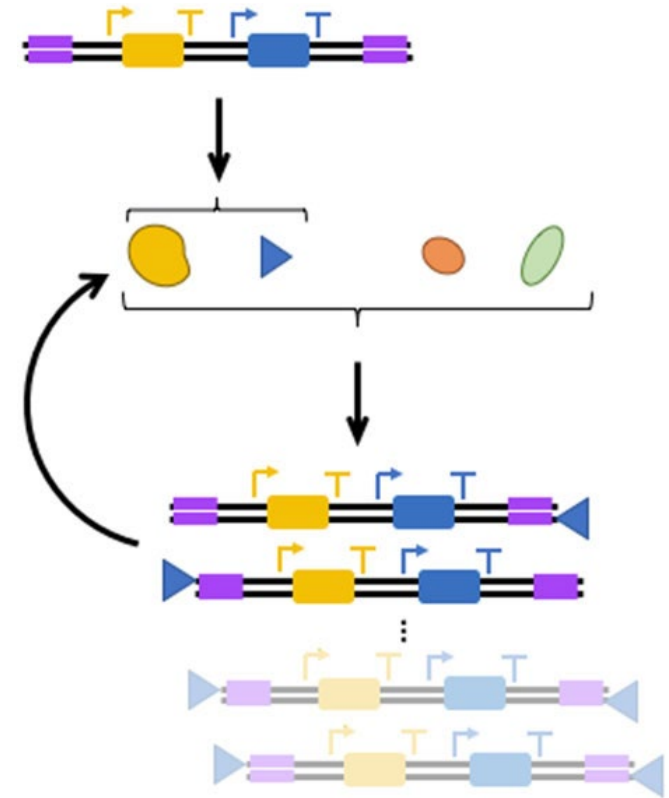
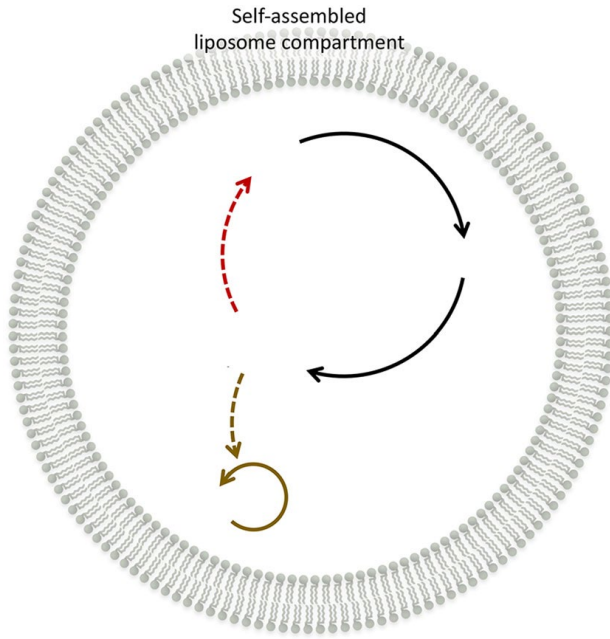
Bottom-up design, characterization and implementation of gene regulatory networks.



Vesicles and communication



DNA replication



Towards an artificial cell

