Green Revolution - SYNTAX
Establishing the fastest growing phototrophic organism as a chassis for synthetic biology

We established Synechococcus elongatus UTEX 2973 with a world record doubling time of under 80 min, providing a fast growing and easy to handle phototrophic chassis. For that, we restore its natural competence, establish the CRISPR/Cas12a system for multiplexed genome engineering and enable the utilization of plasmids as a tool for rapid design testing. Furthermore, we expand last year’s Golden Gate based MoClo toolbox and accelerate the complete cloning workflow by automated plating, colony picking and plasmid purification using the Opentrons OT-2. By providing the fastest phototrophic chassis to the community, we like to pave the way for phototrophic organisms in Green Synthetic Biology.

Growth optimization
Test different parameter to identify optimal growth conditions. To accelerate this process we fed the data to a polynomial regression model.

Standardization & Measurement
Dependence between Growth and Expression Rates
- Missing correlation between light intensity and expression rates
- Tracking of expression on single cell level via FACS analysis
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Light
- Light intensities in incubator are inhomogeneous
  Construction of light intensity distribution model

Media
- No established standardized recipe for BG11 media in the cyanobacterial community
  Evaluation of all provided media

Strain Engineering
Plasmid curing
- Genome (1.1Mbp)
- pANS
- pPIL
- Antibiotics
- pPilN cured
- No Antibiotics
- pPilN cured
- Final cured strain

Easy Transformation
- Nonsense point mutation in the pPilN gene of UTEX
  Restored natural competence by reversing mutation

Genetic Manipulation
- CRISPR/Cas12a system for multiplexed genome engineering
- Enable the community to edit the genome

Lab Automation
Transformation
- Temperature module for heat shock

Plasmid purification
- Capture plasmid

Phage t7
- We provide a low budget solution for fully automated cloning
  - Colony Picking system (CPU) below 300 US$ per day
  - Our hardware and software solutions are open source and available via GitHub

Integrated Human Practice

Acceptance for Green Synthetic Biology

Golden Gate Collaboration
Our team set the goal to introduce other iGEM teams to Golden Gate Cloning.

Interlab
Comparing different Assembly Protocols

Exchange
Automation
Transfer Protocols

Acceptance
I want to test and regulate many different genes - would that be possible if I would use your cloning system?

Integrated Human Practice

"In my opinion, a good terminator has even more effect than a strong promoter"

"My major bottleneck is colony picking"

"In my opinion, Green Synthetic Biology is already in society!"