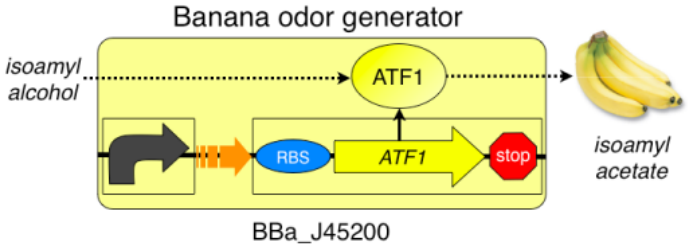
BioBuilding: Synthetic Biology for Students: Eau that Smell

LAB 1: Eau that smell

- Compare 2 competing designs to optimize system performance



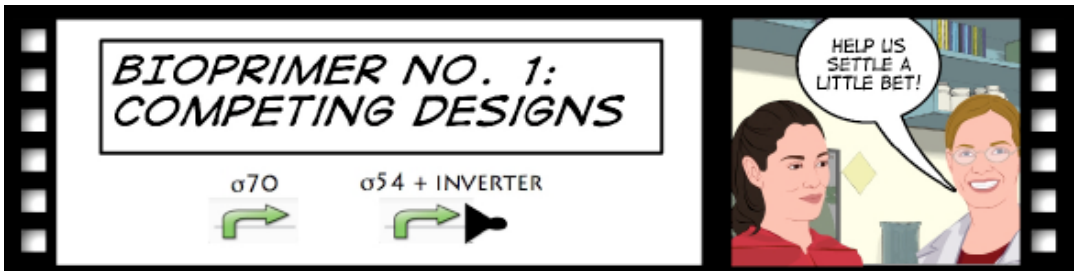
Acknowledgments: This lab was developed with materials and guidance from the MIT 2006 iGEM team, as well as technical insights and help from Ginkgo Bioworks

Objectives

By the conclusion of this laboratory investigation, the student will be able to:

- Explain how synthetic biology as an engineering discipline differs from genetic engineering.
- Explain the population growth curve of bacteria.
- Culture bacteria using proper microbiology methods.
- Measure the growth of a bacterial population.
- Define and properly use synthetic biology terms: Part, Device, Inverter.
- Define and properly use molecular genetics terms: Promoter, ribosome binding site ("RBS"), open reading frame ("ORF"), Terminator, Plasmid.

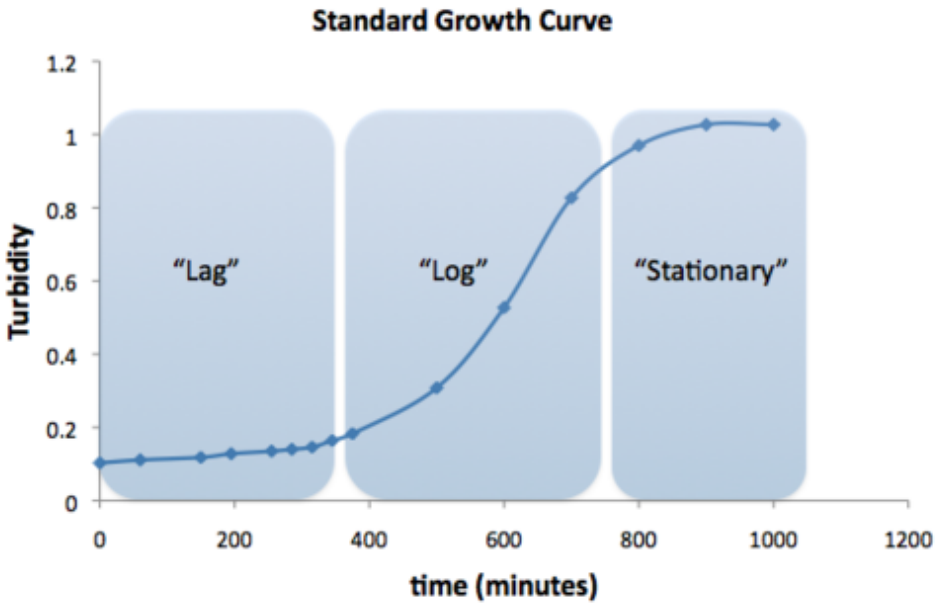
Introduction



For the 2006 iGEM competition, MIT students designed Eau d'coli, *E. coli* that smell like bananas when their population is in the stationary phase. They did this by inserting a device that contains a stationary phase sensitive promoter coupled to a banana smell device, a device that contains a ribosome binding site (RBS), an open reading frame (ORF) that codes for the ATF1 enzyme and terminator sequences. The ATF1 enzyme converts isoamyl alcohol to isoamyl acetate, the molecule that gives bananas their characteristic smell.

It has been suggested that a device to generate the banana smell during the bacteria's log (or exponential) phase of population growth will be helpful. There are two ways to accomplish this. Both approaches will continue to use the original

banana smell device but alter the function of the promoter. One approach couples the banana smell device to a new part, a log phase promoter. The other approach uses the same promoter but adds a genetic inverter. Synthetic biologists have constructed these devices for us and transformed bacteria with them.



We have been sent four strains of *E. coli.* Each contains a different device:

Sample 1-1. The original Eau d'coli device
Sample 1-2. The original Eau d'coli device but with an inverter added between the promoter and the RBS.
Sample 1-3. The banana smell generator coupled to the log phase promoter
Sample 1-4. A strain of *E. coli* that has no smell generating devices.

Our task will be to grow these bacterial populations and test for the banana smell as the population moves through the log phase and into the stationary phase. We will determine the population growth by using a spectrophotometer or the McFarland Turbidity Standards to measure the density of the bacteria in liquid culture. As the population increases we can assess the increasing banana smell, comparing the smell to dilutions of banana extract.

Procedure

There are two versions of this lab. Your teacher will inform you of the one you will be doing. Click on the links below to go to the detailed procedure:
Protocol A

Protocol B

When you've finished your experiments (Protocol A or Protocol B), upload your data to the BioBuilder site that's here. You'll be able to compare what you've measured to what other BioBuilders around the country have seen.

Lab Report

As you write, be sure to define and properly use all highlighted terms throughout the introduction and other parts of the lab.

I. Introduction

- Provide a brief introduction describing the field of synthetic biology.
- Briefly describe the purpose of the lab. What are we trying to do here?
- Explain how the banana smell generator functions.
- Why are we using optical density to measure the population?
- Explain each phase of the bacterial population growth curve.
- Presume that a reader of your lab report has not read the assignment.

II. Methods

- You do not have to rewrite the procedure.
- Explain why you did each step of the protocol.

III. Results

- Present the data tables in clear format.
- Draw population growth curves of the class mean data for each sample. Indicate on each curve when you could smell bananas.

IV. Discussion

- Describe the results: Were we able to measure the population growth? Were we able to smell bananas? Did each device produce the same results? Did the genetic systems affect the growth curve of the bacteria? Explain your answers.
- Analyze the data: Be sure to discuss how each part of the experiment adds to your conclusion.
- Discuss errors and other reasons for data variability.
- How confident are you in the results? Are you equally confident in both the growth data and the smell data? Explain.
- Is using smell to measure the banana smell valid? Why or why not?
- What did methods did you use to try to increase your confidence in the results?
- How might we try to change this system so that we can quantify the banana smell? Would we be better off using a different kind of signal? If so, what would you suggest?
- If you could construct a different genetic system, what might you construct? What would you need to do?

V. Citations and references

- Be sure these are of good quality.
- Embed citations.
- Follow proper reference format.