IPRO 302 SYNTHETIC BIOLOGY
Project plan S06

Introduction

GENERAL IPRO PLAN:

The overall general plan for this IPRO and every IPRO is to improve our team work, project management and peer evaluation skills.

SPECIFIC IPRO PLAN:

The specific plan for our IPRO is construct a synthetic biological circuit, that is both novel, aesthetically pleasing and independent of the host organism. This is a first step toward implementing entirely synthetic organisms that can be designed to a specification.

The target we have chosen is a synchronized biological oscillator, implemented in a vertebrate organism, the zebrafish. To produce an organism requires a wide variety of skills and tasks. Accordingly we have organized into subteams, each responsible for progress in some area, with an administrative sub team overseeing all the events. The sub teams with their basic functions are-

FISH BREEDING

This group is responsible for breeding the fish, feeding the fish, cleaning out the tank, and developing protocols for transferring the circuit into fish to create transgeneic organisms

CLONING

The Cloning sub team goals are to design and construct the physical incarnations of our genetic systems – the physical pieces of DNA encoding our system. cloning the fish, taking care of the fish cell culture, and creating a cloning vector that can be inserted into the fish.

ADMINISTRATION

The administration team coordinates the activities of the subteams as a whole and produces team-wide products, such as documentation and IPRO day materials. It is constituted of the subteam leaders and other coordinating personnel.

FUTURES

The futures team is repsonsible for the creative development of the project plan as a whole – this includes what the team can, should, and might do in the future.

MODELING

The modeling team will focus on the development of computational and mathematical models of pathways function to facilitate design of appropriate genetic elements.

subteam plans:

ADMINISTRATION

One major role of the administration team includes coordination and overseeing of the subteams.

documentation

Documentation includes specific protocols, keeping records of what have been done and either creating a new website or continually updating the previous website.

Ipro Deliverables

The adminstration also keeps track of deliverables such as plans, reports, and the IPRO day material.

FISH

General

The general tasks of the Fish subteam include cleaning the lab, developing wild-type zebra fish colonies, purchasing fish and purchasing fish tanks.

Develop husbandry protocols

We need to work out a good feeding and cleaning schedule, and continually research more into breeding and upkeep.

Develop methods to clone into fish

Research needs to be done about ways of cloning into the fish, and then determined if the method chosen is viable with our money supplly.

Produce embryos/ breed fish

Developing of techniques to prepare the brine shrimp and developing of methods for breeding need to be done. After this we can possible breed the goldfish.

CLONING

Construction

This subteam will work on constructing Tet-YFP, Tet-CFP, along side adding the Zeo into the pUHD-GFP. This subteam will also need to determine source and specific version of RFP, and then to construct Tet-RFP. Currently our system uses Tet-ON, this subteam will also get Tet-OFF working.

Design

This subteam is responsible for the design modifying and porting the oscillator plasmid, suitable for the eukaryotic cells. A part of this is to look for the mammalian systems that will better suit the fish rather than the system used in bacteria.

3 promoter/repressor pairs will be needed for our system, (i.e. TET system), and this team will research a way to put them together in a workable fashion. Some possible candidates for this are: LexAse; CMViptg(CMV-cytomegalovirus , viral promoter); Gal4. Also this subteam will investigate some possible Pisces specific promoters.

They may also look into siRNA systems as an alternative way to our current system.

They will then develop a specific assembly plan, which will be worked on.

Cell culture

This subteam will research to determine the Transfection protocol, which is a method of insertion of plasmids into cells. Since the cell the team will be working on this semester are eukaryotic, which are larger and more complex than the bacterial cell it will require a different procedure for Transfection, than the bacterial Transfection. Along side this they will also determine the kill curves of Zeo gene and G418 genes.

A possible way for Transfection is the to utilize the liposomes which will be researched by this team. They will also work to generate rtTA (reverse tetracyline-dependent transactivator) cell line (with neomycin/G418 resistance???)

They will also make second reporter (GFP/YFP/CFP/RFP) LINE (Zeomycin resistant), and unregulated XFP vectors as control?

MODELING

Optimization

Target

Convert present code to another faster language such as C++, Java, or work out a way to make our present Matlab coding faster.

Possible target (study and determine feasibility)

-Possibly look into making our program web compatible.

-Our gui file needs to be tinkered with a little to create more compatibility.

Elaboration

Target

The current code uses two cells, but we need to convert the language to work for N number of cells, and get these cells synchronized.

Possible target (study and determine feasibility)

We need to improve our analysis tools, using analyses such as nonlinear least squares that will search for an optimum output.

FUTURES/DESIGN

Several items that the futures/design subteam will look into include, what has been done in Synthetic biology, developing possible attractive and feasible targets fro further development and conducting biological feasibility analysis.