

PIPECLEANER PLASMIDS

INSTRUCTOR GUIDE

Purpose

This activity will expose students to the real life applications of synthetic biology and catalyze creative thinking for future, potential iGEM projects.

Materials for each group (groups of 3 to 5)

- Pipe cleaner
- Gene bead set (12 colours)
- Promoter, RBS, Terminator bead set (3 colors)
- Scenarios
- Gene Legend

Instructions

Provide some background information:

- How is the DNA of E. coli different?
- What are plasmids and how can they be created?
- What is synthetic biology?
- What are some of the controversies of using synthetic biology?

To start, have group choose a scenario and let them create their ideal plasmid with no size limit. The students must find the important information from the scenarios to choose the appropriate genes for their plasmids.

Then, impose size restrictions on the plasmids based on instability of larger size plasmids. The new size to aim for is around 25kb. The students will then have to choose what genes are the most beneficial to their given scenario and which ones can be eliminated or changed.

Swap plasmids with another group and try to guess which scenario the plasmid belongs to. Allow the groups to give feedback and suggestions to each other.

Finally, have the groups present their plasmid along with justification for why they chose the certain genes.

Optional:

- Introduce new factors to the scenario as the students are creating their plasmids.

Example: “There have also been traces of lead/base/acid found in _____”

- Have the students create their own scenario using the gene legend as a resource, or allow them to add new genes to the legend. Be creative! It could be imaginary!

PIPECLEANER PLASMIDS SCENARIOS

1. The Great Pacific Garbage Patch

<http://www.youtube.com/watch?v=2VrrxMliwgQ>

This ocean landfill is a gyre of marine litter in the central North Pacific Ocean. It was formed gradually as a result of marine pollution gathered by oceanic currents. Unlike debris, which biodegrades, the photodegraded plastic disintegrates into ever smaller pieces while remaining a polymer. Let's imagine that the plastic composition of the garbage is 50% plastic bottles (which photodegrades into polymer A), 35% plastic bags (polymer B) and 15% can be attributed to plastic action figures (polymer C). Some of these plastics decompose within a year of entering the water, leaching potentially toxic chemicals so time-sensitive cleanup is important. The conditions are cold (2 to 12°C) saltwater, with plastic particles spanning over a large area with a variable composition.

GARBAGE PATCH

NAME	FUNCTION	SIZE	COLOR
MPA_8	Plastic container (polymer A) metabolism	6	PURPLE
MPB_4	Plastic bag (polymer B) metabolism	3	PURPLE
MPC_3	Plastic toy (polymer C) metabolism	2	PURPLE
MLT_2_5	Suitable for life in temperature range from 2 to 12°C	5	BROWN
MHDE_3_4	Polycarbon Digestion – Necessary for ALL Polymer metabolism	3	ORANGE
MOME_6	Oxygen Metabolism – Necessary for ALL organisms	3	WHITE
REPRODUCTION			
RHRR_7	High Reproduction Rate – Grows quickly	7	OLIVE
RMSC_5	Medium Colony Size	5	LTBLUE
SURVIVAL			
SDRE_7	Salinity survival	5	GRAY
OSG-ISG	Suicide Gene – Inducible Suicide BioDesignOrganismide	5	DKBLUE
OSFTR	Fluorescent Tracking – Advantage easy & cheap	1	MAROON
SUM			45

2. Gulf of Mexico Oil Spill

A drilling rig has collapsed off the coast of Florida in the Gulf of Mexico. It is estimated that it will spill up to 1 billion barrels of crude oil before the leak is sealed. The large size of this disaster will require a large remediation effort in order to restore the water to a clean state. Of note for bioengineering strategies, the climate in the water requires bacteria, which can thrive in warm waters (20°C – 28°C) as well as process crude unrefined oil as a substrate. The spill is quite close to shore, so it is necessary to rapidly clear the product (within 6 months) before it contaminates coastal ecosystems.

GULF OIL SPILL

METABOLISM			
MPF_1_6	Pure Fuel - Crude Unrefined Oil Metabolism	5	BLACK
MHT_2_7	Suitable for life in temperature range from 22 to 30°C	6	BROWN
MHDE_3_4	Hydrocarbon Digestion – Necessary for ALL Fuel metabolism	6	ORANGE
MOME_6	Oxygen Metabolism – Necessary for ALL organisms	3	WHITE
REPRODUCTION			
RHRR_7	High Reproduction Rate – Grows quickly	7	OLIVE
RMSC_5	Medium Colony Size	5	LTBLUE
SURVIVAL			
SDRE_7	Salinity survival	5	GRAY
OTHER			
OSGHT_6M_3	Suicide Gene – High Telomerase – Survive 6 months	3	DKBLUE
OSFTR	Fluorescent Tracking – Advantage easy & cheap	1	MAROON
SUM			41

3. Landfill

A landfill is a method of solid waste disposal by burying it under layers of earth. In Canada, we have a recycling system accessible to 96% of Canadians; however, there are some plastics which end up in the landfill since they are considered a contaminant in residential recycling programs. We propose using synthetic biology to help degrade the unrecyclable plastic products. The composition of the disposed plastic is: 25% low density polyethylene frozen food bags (polymer B), 50% containers (polymer A and B), which previously contained crude motor oil and still has oil residues, and 25% Polystyrene toys (polymer C). The landfill is large and is of a mid temperature range: 13oC – 22oC.

LANDFILL

METABOLISM			
MPA_8	Plastic container (polymer A) metabolism	6	PURPLE
MPB_4	Plastic bag (polymer B) metabolism	3	PURPLE
MPC_3	Plastic toy (polymer C) metabolism	2	PURPLE
MPF_1_6	Pure Fuel - Crude Unrefined Oil Metabolism	5	BLACK
MMT_2_3	Suitable for life in temperature range from 10 to 24°C	3	BROWN
MHDE_3_4	Hydrocarbon Digestion – Necessary for ALL Fuel metabolism	6	ORANGE
MHDE_3_4	Polycarbon Digestion – Necessary for ALL Polymer metabolism	3	ORANGE
MOME_6	Oxygen Metabolism – Necessary for ALL organisms	3	WHITE
REPRODUCTION			
RHRR_7	High Reproduction Rate – Grows quickly	7	OLIVE
RMSC_5	Medium Colony Size	5	LTBLUE
SURVIVAL			
OTHER			
OSG-ISG	Suicide Gene – Inducible Suicide BioDesignOrganismide	5	DKBLUE
OSFTR	Fluorescent Tracking – Advantage easy & cheap	1	MAROON
SUM			49

PIPECLEANER PLASMIDS GENE LEGEND

NAME	FUNCTION	SIZE	COLOR	Example	Scenario
METABOLISM					
MPA_8	Plastic container (polymer A) metabolism	6	PURPLE		
MPB_4	Plastic bag (polymer B) metabolism	3	PURPLE		
MPC_3	Plastic toy (polymer C) metabolism	2	PURPLE		
MPF_1_6	Pure Fuel - Crude Unrefined Oil Metabolism	5	BLACK		
MHM_1_8	Heavy metals (Lead)	6	BEIGE		
MHT_2_7	Suitable for life in temperature range from 22 to 30°C	6	BROWN		
MMT_2_3	Suitable for life in temperature range from 10 to 24°C	3	BROWN		
MLT_2_5	Suitable for life in temperature range from 2 to 12°C	5	BROWN		
MHDE_3_4	Hydrocarbon Digestion – Necessary for ALL Fuel metabolism	6	ORANGE		
MHDE_3_4	Polycarbon Digestion – Necessary for ALL Polymer metabolism	3	ORANGE		
MOME_6	Oxygen Metabolism – Necessary for ALL organisms	3	WHITE		
REPRODUCTION					
RHRR_7	High Reproduction Rate – Grows quickly	7	OLIVE		
RMRR_5	Medium Reproduction Rate – Grows moderately	5	OLIVE		
RMSC_5	Medium Colony Size	5	LTBLUE		
RSCS_4	Small Colony Size	4	LTBLUE		
SURVIVAL					
SDRE_7	Salinity survival	5	GRAY		
SAAPS_4	Acidic pH Survival	4	PINK		
SBPS_4	Basic pH Survival	6	PINK		
OTHER					
OSGHT_6M_3	Suicide Gene – High Telomerase – Survive 6 months	3	DKBLUE		
OSG-ISG	Suicide Gene – Inducible Suicide BioDesignOrganismide	5	DKBLUE		
OSFTR	Fluorescent Tracking – Advantage easy & cheap	1	MAROON		

*Every gene requires a Promoter (Green), Ribosome binding site (Yellow) and Terminator (Red) bead.