

COLOR CODING KEY

Spectrophotometry Measurements

Construct Group

Plasmid Group

Interlab

Cell Culture/Plating

Biobrick Group

Cyanobacteria Transformation Group

Experimental Verification

Plasmid & Construct Design Group

Week 18

October 1, 2018

Biobrick Group (Natalie/Matt)

- RE digest of backbone
- PCR of psbA2, cpc560, idiA, cscB
- PCR purification of PCR products
- RE digest of PCR products

Experimental Verification Group (Karthik/Elon)

- Luciferase expression after 1h high light exposure of white light
 - Pipetted 95 μ L of cells onto half of the wells on the plate
 - High light exposure for an hour in the incubator
 - Afterwards, pipetted 95 μ L of cells not exposed to high light for 1 hour
 - Added 5 μ L of decanal to all wells
 - Parafilmed well plates
- Rows 1-6 High light exposure, 7-12 no high light
- A:WT, B-C: cpc2#1,3#1, D-E: cpc 2#1,3#1, F-H: psbA2 1-3

0	4	0	0	4	6	0	0	0	2	0	0
203	935	1331	1032	634	802	473	337	416	387	228	220
119	521	768	519	207	157	496	142	130	107	170	107
165	232	186	201	247	157	136	138	98	153	167	134
274	952	561	287	396	454	314	391	310	308	251	228
4	4	0	8	2	0	2	2	2	0	0	2
0	2	0	2	4	2	4	2	2	0	2	0
0	6	2	4	2	2	2	0	0	0	2	0

October 2, 2018

Biobrick Group (Natalie/Matt/Karthik/Lin)

- Ligation of PCR products and backbone

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- Transformation and plating of products cpc560, idiA, psbA2, cscB
- RE digest of cpc560, idiA, psbA2, cscB again
- Nanodrop of RE digest
 - cscB = 9.9 ng/ μ L, 20.9 ng/ μ L, 21.1 ng/ μ L
 - idiA = 36.9 ng/ μ L
 - psbA2 = 51.3 ng/ μ L
 - Cpc 560 = 39.4 ng/ μ L

October 3, 2018

Biobrick Group (Natalie/Matt/Karthik)

- Transformation of cpc560, idiA, psbA2, cscB

October 4, 2018

Biobrick Group (Priya/Woody/Sara)

- Inoculation of plates from yesterday
- Miniprep of inoculation and sequencing

Experimental Verification (Elon)

Water	0	0	0
Glucose Control	0.996	0.958	0.965
Starch Acetate	0.078	0.049	0.066
Starch Fructosidase	0.145	0.141	0.149
1 Acetate	0.001	0.003	-0.008
1 Fructosidase	-0.014	-0.015	-0.019
2 Acetate	-0.021	-0.020	-0.023
2 Fructosidase	-0.017	-0.018	0.094
3 Acetate	-0.020	-0.011	-0.017

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3 Fructosidase	-0.009	0.000	-0.009
4 Acetate	-0.004	0.003	-0.003
4 Fructosidase	-0.009	-0.007	2.490
5 Acetate	0.011	0.007	0.027
5 Fructosidase	0.010	0.018	0.012
6 Acetate	0.006	0.003	0.002
6 Fructosidase	-0.005	-0.010	-0.002
7 Acetate	0.004	-0.005	0.025
7 Fructosidase	N/A	N/A	N/A
8 Acetate	-0.011	0	-0.015
8 Fructosidase	-0.005	-0.031	0.014
9 Acetate	-0.016	-0.006	-0.031
9 Fructosidase	0.000	0.043	-0.008

*Key:

#1 = cscB #3 150 mM NACL 1 mM IPTG 10 uM Spec 10 uM Strep Room Temp

#2 = cscB #2 150 mM NACL 1 mM IPTG 10 uM Spec 10 uM Strep Room Temp

#3 = cscB #2 No Salt NACL 1 mM IPTG Incubator

#4 = cscB #2 150 mM NACL 1 mM IPTG Incubator

#5 = cscB #2 150 mM NACL Incubator

#6 = cscB #4 #1 150 mM NACL 1 mM IPTG 10 uM Spec 10 uM Strep Incubator

#7 = cscB #4 150 mM NACL 1 mM IPTG 10 uM Spec 10 uM Strep Incubator

#8 = cscB #2 150 mM NACL 1 mM IPTG 10 uM Spec 10 uM Strep Incubator

#9 = cscB #4 #2 NACL 150 mM 1 mM IPTG Incubator

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Analyte	Analyte (g/L)
Starch	0.033
Glucose	
Starch	0.080
Sucrose	
1 Glucose	0.001
1 Sucrose	-0.002
2 Glucose	0.000
2 Sucrose	0.046
3 Glucose	0.000
3 Sucrose	0.000
4 Glucose	0.001
4 Sucrose	-0.001
5 Glucose	0.010
5 Sucrose	-0.004
6 Glucose	0.002
6 Sucrose	-0.004
7 Glucose	0.006
7 Sucrose	-0.012

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8 Glucose	0.000
8 Sucrose	0.007
9 Glucose	0.000
9 Sucrose	0.021