

Names of each student: Elizabeth Plumart

Date: 09.18.23

Protocols Performed: Heat shock

Purpose: Transform phaB, phaC, pChlamdy into E. coli

Results: Success

Conclusions/Next Lab goals: Overnight culture to make glycerol stocks, transform phaA, tdTomato, algae promoter, algae terminator into E. coli

Extra notes:

Names of each student: Elizabeth Plumart, Chloe Benjamin

Date: 09.22

Protocols Performed: Overnight culture

Purpose: Grow up colonies from transformation (phaB, phaC, pChlamdy)

Results: **Success**

Conclusions/Next Lab goals: Mini-prep

Extra notes:

Names of each student: Elizabeth Plumart

Subteam name: Algae PHB

Date: 09.23

Protocols Performed: Mini-prep

Purpose: Extract plasmid (phaB, phaC, pChlamdy) from overnight culture

Results: Success

Plasmid/Gene, Colony	Concentration (ng/ul)	260/280
pChlamdy, 1	47.8	1.92
pChlamdy, 2	56.4	1.92
pChlamdy, 3	48.7	1.93
phaC, 1	37.9	1.86
phaC, 2	51.5	1.90
phaC, 3	64.4	1.94
phaB, 1	505.7 (measured 3 times)	1.85 (260 = 10.114, 280 = 5.464)
phaB, 2	48.3	1.93
phaB, 3	48.6	1.91

Conclusions/Next Lab goals: Transform kit plate parts, restriction digest & ligation to build expression construct

Extra notes:

Names of each student: Elizabeth Plumart, Chloe Benjamin

Date: 09.25.23

Protocols Performed: Heat shock

Purpose: Transform phaA, CaMV 35S, 3' UTR, mCherry into E. coli

Results: Success

Conclusions/Next Lab goals: Overnight culture to make glycerol stocks, colony PCR on kit plate parts

Extra notes:

Names of each student: Chloe Benjamin

Date: 09.26.23

Protocols Performed: overnight culture of E.coli plates (phaA, mCherry, CaMV 35S, 3' UTR)

Purpose: to prepare for the Miniprep and making glycerol stocks tomorrow

Results: Success

Conclusions/Next Lab goals: Colony PCR, Miniprep, Glycerol stocks, Restriction Digest to create algae construct, Ligation, Transform algae construct into E.coli

Extra notes: It was decided that the colony pcr will be done on Wednesday, 9/27/23 instead. The shaker in nsc 318 was not working last night, so I put the overnight culture tubes in another incubator in nsc 318. The E.Coli plates were placed back in the fridge in nsc 110 and the purple box containing antibiotics were placed in the freezer in nsc 110.

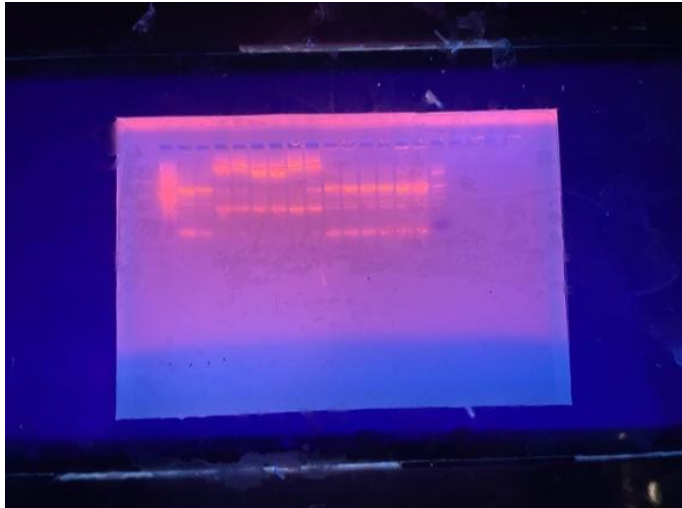
Names of each student: Xucheng Zhang

Date: 09.27.23

Protocols Performed: colony pcr and gel electrophoresis

Purpose: confirms E.coli has the genes mCherry (reporter gene), CaMV 35s(promoter), and 3' UTR (terminator)

Results: Success



Conclusions/Next Lab goals: Miniprep

Extra notes:

Names of each student: Chloe Benjamin, Elizabeth Plumart

Date: 09.27.23

Protocols Performed: miniprep

Results: Success

Plasmid/Gene, Colony	Concentration (ng/ul)	260/280
phaA 1	80.4	1.92
phaA 2	96.8	1.93
phaA 3	90.3	1.92
CaMV 1	52.5	1.98
CaMV 2	57.1	1.98
CaMV 3	71.1	1.92
3' UTR 1	71.5	1.95
3' UTR 2	62.3	1.95
3' UTR 3	57.3	1.96
mCherry 1	46.6	2.01
mCherry 2	65.3	1.99
mCherry 3	61.6	1.99

Conclusions/Next Lab goals: Miniprep

Extra notes:

Names of each student: John Biggs

Date: 09.28.23

Protocols Performed: Restriction digest

Restriction digest enzymes

Gene	Enzymes
CaMV 35S (algae promoter)	SpeI & PstI
mCherry (reporter)	XbaI & PstI
phaA	XbaI & PstI
phaB	XbaI & PstI
phaC	XbaI & PstI

Purpose: Prepare construct for expression

Results: phaA, phaB, phaC success, CaMV35S, mCherry fail

Conclusions/Next Lab goals:

- The next step is completing the remaining restriction digests, gel extractions, and ligations to finish building the construct we want to express.

Extra notes:

Names of each student: Chloe Benjamin, Elizabeth Plumart

Date: 10/2/23

Protocols Performed:

- Gel electrophoresis to verify if the restriction digests completed on 9/28/23 were done successfully

Restriction digest enzymes

Gene	Enzymes
CaMV 35S (algae promoter)	SpeI & PstI
mCherry (reporter)	XbaI & PstI
phaA	XbaI & PstI
phaB	XbaI & PstI
phaC	XbaI & PstI

Run out on gel, gel extraction on band.

Purpose: to prepare for construct for expression

Supplies used/Materials:

Results: The DNA bands did not travel far enough down the gel, so we did not get successful results to analyze.

Conclusions/Next Lab goals: The next goal is to repeat the restriction digests on 9/28/23.

Extra notes: