

Name: Katie Fitten

Date: 7/22/14

WEEKLY ONE SENTENCE:

Continue testing RBS primers from ote10

DAILY ONE SENTENCE:

clone RBS primers into mcherry

METHOD(S) OF CHOICE:

PCR

PCR purification
gel electrophoresis

EXPECTED RESULT:

band @ 1 kb

☒ check if APE file exists on Drive)

TEMPLATE(S): mcherry

OLIGO(S): 74-78, 363

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE: 362

NEGATIVE:

w/o p04mcherry

CONFIRMATION?

True

GEL LANE ORDER:

GEL PICTURE / OTHER SPACE:

Name: Jennifer Zhang

Date: 7/20/14

WEEKLY ONE SENTENCER: combine transcription
ZBs and promoter primers

DAILY ONE SENTENCER: grow up colonies from transformations
positive group presentation

METHOD(S) OF CHOICE:
cultures (LB + CM)

EXPECTED RESULT:
cultures to be manipulated tomorrow

☒ check if APE file exists on Drive

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

CONFIRMATION?

GEL LANE ORDER:

GEL PICTURE / OTHER SPACE:

Name: Jennifer Zhang

Date: 7/25/14

WEEKLY ONE SENTENCE: continue from BLA subcloning
RB5 and promoter pairs

DAILY ONE SENTENCE: - miniprep RHZ, RH1 then send for
promoter PCR w/ RB5
digest / ligate then transform

METHOD(S) OF CHOICE:
miniprep
PCR

digest / ligate
transformation

EXPECTED RESULT:
colonies on LB + CM plates
band ~ 1 kb

☒ check if APE file exists on Drive)

TEMPLATE(S): RB5 PL12

OLIGO(S): 80-85, 303

THERMOCYCLER SETTINGS: 94-2' [94'-15", 60-15", 68-1.5'] x 31

CONTROLS: UAC-4', 4°C

POSITIVE: 302

NEGATIVE: no template

CONFIRMATION?

81

GEL LANE ORDER:

1 kb
RH1 PL1
RH2 RH1
RH1 RH1
RH1 RH1
RH1 PL1
RH1 PL1

GEL PICTURE / OTHER SPACE:

Name: Troy von Beck

Date: 8/5/2014

WEEKLY ONE SENTENCER:

Finish formatting the wikis Notebook

DAILY ONE SENTENCER:

Transform selected promoters from Brobrck plates

METHOD(S) OF CHOICE:

Heat shock
Em Antibiotic selection

EXPECTED RESULT:

Colonies with useful BioBricks

☐ check if APE file exists on Drive)

TEMPLATE(S): To be determined

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

DH5a untransformed

Which promoters?
Andersun Promoters

Part #
BBa_J23100
BBa_J23102
BBa_J23101



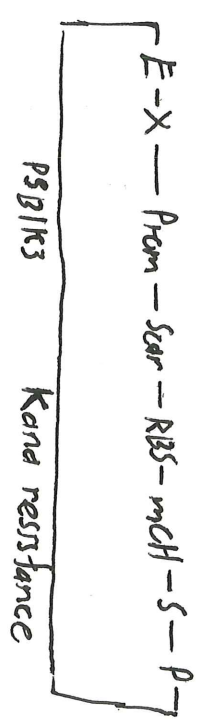
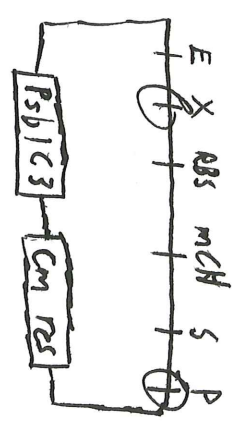
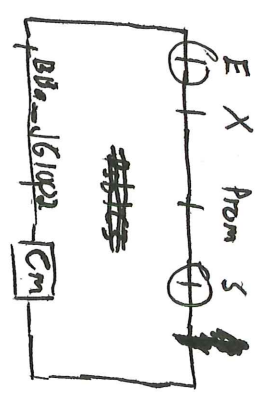
BBa_J04450

CONFIRMATION?

GEL LANE ORDER:

GEL PICTURE / OTHER SPACE:

Strength	Location
1	2014 Kd Plate 4, well 17D
.86	" " " well 17H
.7	" " " well 17F



Name: M. S. C.

Date: 8/6/14

CONFIRMATION?

Tube gel

WEEKLY ONE SENTENCER:

- work on cloning ees into α , β & γ

- cloning ees into smuB

DAILY ONE SENTENCER:

- clone smuB, smuB, α -1, β & γ into lactate
plasmid

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

- digestion (E & P), ligation, quick gel, gel extraction & purification

EXPECTED RESULT:

Bands @

1600 bp for α -1 & 554 bp for γ subunit

467 bp " smuB

1088 bp " smuB Δ Att-transformed

1211 bp " B subunit colonies on Gm plates

☒ check if APE file exists on Drive

TEMPLATE(S): mCherry plasmid

OLIGO(S): N/A

THERMOCYCLER SETTINGS: Digest 37 (1hr @ 37°C)

CONTROLS:

POSITIVE:

NEGATIVE:

H₂O without inserts. w/ vector

Name: Jennifer Zhang

Date: 8/11/14

WEEKLY ONE SENTENCE: put proteins B,C in Diels Alder format ✓

DAILY ONE SENTENCE: digest protein B,C PCR product, ligate with pG7-1C3 backbone, transform ✓

METHOD(S) OF CHOICE:

digest / ligate w/ E/P ✓
PCR purify
gel extract
transformation
EXPECTED RESULT:
 α - 1.5 kb ✓
mercury - 2 kb ✓

(X) check if APE file exists on Drive)

TEMPLATE(S): α , B, Y, B, C

OLIGO(S):

THERMOCYCLER SETTINGS: 37°C 1 hr
80°C 20 min ✓

CONTROLS:

POSITIVE:

NEGATIVE:

CONFIRMATION?

8-1

GEL LANE ORDER:

1 kb α mch ✓

GEL PICTURE / OTHER SPACE:

(805) 1000's M₀₀₀ = (X) result 1.75

Name: Amirullah Toshi

CONFIRMATION?

Date: 8/12/14

WEEKLY ONE SENTENCER:

Inserting ^{genes} α , β , γ , B and C into

PSB-1C3

GEL LANE ORDER:

DAILY ONE SENTENCER:

Digest genes α , β , γ , B, C and PSB-1C3
backbone

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

EXPECTED RESULT:

☐ check if APE file exists on Drive)

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

Name: Jennifer Zhang

Date: 8/14/14

WEEKLY ONE SENTENCE: continue trying to put genes α , β , γ , B, C into λ B-1C3

DAILY ONE SENTENCE: digest α , β , γ , B, C, insertory @ E/P

ligating and transforming

mini-prep β , γ , B, C

METHOD(S) OF CHOICE:

digest E/P

gel extraction

ligation

transformation

EXPECTED RESULT:

α \rightarrow 1600 bp

β \rightarrow 1200 bp

γ \rightarrow 550 bp

B \rightarrow 460 bp

C \rightarrow 1088 bp

insert \rightarrow 2 kb

check if APE file exists on Drive

TEMPLATE(S): α , β , γ , B, C, insertory

OLIGO(S):

THERMOCYCLER SETTINGS:

57°C @ 1 hr

60°C @ 20 min

CONTROLS:

POSITIVE:

N/A

NEGATIVE:

N/A

CONFIRMATION?

8-1

GEL LANE ORDER:

1 kb α β γ B C insert

GEL PICTURE / OTHER SPACE:

contamination \rightarrow threw out products

Name: Mishi.C

CONFIRMATION?

Gel

Date: 8/14/14

WEEKLY ONE SENTENCER:

- Continue trying to insert all genes into the pSB1C2

GEL LANE ORDER:

1kb α β γ δ ϵ mc

DAILY ONE SENTENCER:

- Miniprep cultures of α , β , γ , δ , ϵ

- Digest, ligate & transform genes

- Digestion (ϵ & δ)

- Gel extraction + purification

- Ligation (instant)

- Transformation

EXPECTED RESULT:

- Bands @

1600bp for α
220 for mc

1211 for β
554 for γ

Transformed colonies

1167 for δ
1088 for ϵ

(☒ check if APE file exists on Drive)

TEMPLATE(S):

mCherry

OLIGO(S):

THERMOCYCLER SETTINGS:

Digest with 80°C @ 20 min

CONTROLS:

N/A

POSITIVE:

NEGATIVE:

N/A

GEL PICTURE / OTHER SPACE:

How did this turn out?

Name: Troy

CONFIRMATION?

Date: 8/14/2014

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

Ligate iGEM Anderson Promoters with our RBS machinery

DAILY ONE SENTENCER:

METHOD(S) OF CHOICE:

Digest RH2 machinery X + P

Digest BBA-J23 (100-102) 5 + P

ligase extract RH2 machinery insert + BBA-J2310N backbones

EXPECTED RESULT:

Ligate insert w/ backbones
transform + plate on Amp plates

Pink colonies tomorrow
(☐ check if APE file exists on Drive)

TEMPLATE(S): RH2 machinery, BBA-J23100, BBA-J2310, BBA-J23102

OLIGO(S): X, S, P

THERMOCYCLER SETTINGS: Digest

CONTROLS: None

POSITIVE: None

NEGATIVE: Control ~~amp~~ plate

Poor Quality Gel Extraction
Low Yields w/ contamination, did not progress to ligation

Name: Coleen Tran

CONFIRMATION?

Date: 8/14/14

gel

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

working on promote + RBS + melting ✓

1 B C B D A N ✓

DAILY ONE SENTENCER:

confirming sequencing & helping

METHOD(S) OF CHOICE:

Seamless + Tishui

GEL PICTURE / OTHER SPACE:

- PCR

- digest + ligate

EXPECTED RESULT:

(control) B: ~1200 bp

D: 550

C: ~1100 bp

A: 1600 ✓

(☐ check if APE file exists on Drive)

TEMPLATE(S): same genes

OLIGO(S): 20, 91

THERMOCYCLER SETTINGS:

98°C - 30" - { 96°C - 10"; 60°C - 20"; 72°C - 1' } x 31; 72°C - 2'; 4°C

CONTROLS:

POSITIVE:

NEGATIVE:

no template

↳ results?

How did this turn out?

Name: Stefan Tassoulas

CONFIRMATION?

Date: 08/15/2014

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

- clone ~~plasmid~~ into ~~plasmid~~ λ B8 with GS. \checkmark

DAILY ONE SENTENCER:

- Transform λ B8 into PSB163 \checkmark
- host with GS. DH5 α \checkmark

METHOD(S) OF CHOICE:

GEL PICTURE / OTHER SPACE:

- Digestion, PCR machine, Ligation
E+P \checkmark

EXPECTED RESULT:

Transformed λ B8 clones \checkmark

(☐ check if APE file exists on Drive)

TEMPLATE(S): ~~kan~~ pU57-b, pU57-d, TOPO-d

OLIGO(S): N/A

THERMOCYCLER SETTINGS:

Digestion protocol 37 $^{\circ}$ C for 2 hrs, 65 $^{\circ}$ C 20min, 4 $^{\circ}$ C 16 $^{\circ}$ forever
37 $^{\circ}$ C for 1hr, 80 $^{\circ}$ C for 20mins, 4 $^{\circ}$ C forever

CONTROLS:

POSITIVE:

NEGATIVE:

digest Neg control
H₂O + cutsmart + E+P

Name: Cohen Tran

CONFIRMATION?

Date: 8/5/14

gel

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

working on promoter f685 + melting clones

L H86 H87 H89 - L86 L87 L88 RT

DAILY ONE SENTENCER:

- PCR promote primer into R85 + melting clones

METHOD(S) OF CHOICE:

- contact Tim Moss from USF about collets

GEL PICTURE / OTHER SPACE:

140805 Promote + R85 + melting

- PCR

- gel

EXPECTED RESULT:

21kb

☒ check if APE file exists on Drive)

TEMPLATE(S): ~~2000~~ 42, ~~PCR~~ L1

OLIGO(S): ~~360~~, 363, 86-88

THERMOCYCLER SETTINGS:

CONTROLS: 98°C - 30" { 98°C - 10", 60°C - 20", 72°C - 20" } x31; 72°C - 2', 4°C

POSITIVE:

3602

NEGATIVE:

no template ✓

Name: Katie Fitton

Date: 8/5/14

WEEKLY ONE SENTENCER:

CONFIRMATION?
+ⁱⁿ gel of just plasmid before digestion

GEL LANE ORDER:

DAILY ONE SENTENCER:

WORK ON PUTTING SMO genes in biobrick format

transform SMO genes w/ biobrick plasmid

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

digestion (EsiP), gel extract, ^{ligation} transformation

EXPECTED RESULT:

α - 1600 bp

β & γ - 1500 bp

γ & δ - 1600 bp

(☐ check if APE file exists on Drive)

TEMPLATE(S): mcherry ~~plasmid~~ plasmid

OLIGO(S):

THERMOCYCLER SETTINGS: 1 hour @ 37°C

CONTROLS:

POSITIVE:

NEGATIVE:

w/o restriction enzymes (EsiP)

Name: Ms. C

CONFIRMATION?

gel

Date: 8/5/14

WEEKLY ONE SENTENCE:

GEL LANE ORDER:

- Working on cloning pBS into α , β , γ
- Cloning pBS into 311 pUC19

1 2 3 4 5 6 7 8

DAILY ONE SENTENCE:

- Cloning pBS into pUC19, α -1, β , γ
into ligation plasmid.

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

- Digestion, ligation with (E. coli)
↳ using Digest 37

EXPECTED RESULT:

Sample C: 1600 bp for α -1 1 transformed clones
467 bp " same β
1086 bp " same γ
1211 bp for p subunit
554 bp for γ subunit-1

TEMPLATE(S): pUC57- β , pUC57- γ , Telo- α

OLIGO(S): N/A

THERMOCYCLER SETTINGS: Digest 37

CONTROLS:

POSITIVE: N/A

NEGATIVE:

Digested My control of H₂O + extract + E. coli

Name: *Eden Tran*

CONFIRMATION?

Date: *8/6/14*

no agar plates

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

working on promoter + ABS + mCherry

DAILY ONE SENTENCER:

digest + ligate promoter + ABS + mCherry into vector

METHOD(S) OF CHOICE:

digest, ligate, transformation

GEL PICTURE / OTHER SPACE:

EXPECTED RESULT:

vector: ~2 kb

insert: ~1 kb

☐ check if APE file exists on Drive)

TEMPLATE(S): *486-88, 486-88*

OLIGO(S):

THERMOCYCLER SETTINGS: *37°C for 1 hour (digestion)*

CONTROLS:

POSITIVE:

NEGATIVE:

no template

Name: *Youtie Fitten*

Date: *8/16/14*

WEEKLY ONE SENTENCER:

Work on putting Sma genes in b2briC
form

CONFIRMATION?

yes gel

GEL LANE ORDER:

DAILY ONE SENTENCER:

~~genes~~ *genes* w/ b2briC plasmid again
transform Sma

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

digestion (EcoP), gel extract ligation,
1' transformation

EXPECTED RESULT:

~~acid~~ ~~467 bp Sma~~ ~~1028 bp Sma~~
~~1028 bp Sma~~

☐ check if APE file exists on Drive

TEMPLATE(S): *weherry plasmid*

OLIGO(S): *N/A*

THERMOCYCLER SETTINGS: ~~24°C~~ *1 hour* ~~37°C~~

CONTROLS:

POSITIVE:

NEGATIVE:

no instead of DNA w/ vector

CONFIRMATION?

✓

GEL LANE ORDER:

信

1-dephosphatation

(New method) GEL PICTURE / OTHER SPACE:

7

△

- 1 will give assignment

All the ops for 99 full

 \angle

15051

222

1000

1.31 kL/sec

1.81 μL Decaol

1. A pre

2.0. μ Δ

Name: *Chen Tian*

CONFIRMATION?

Date: *8/21/14*

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

working on ~~BB8~~ promoter + ~~BB8~~ promoter + ~~BB8~~ promoter

DAILY ONE SENTENCER:

mini-prep, ~~tray's~~ ~~BB8~~ - Andersen promoter

METHOD(S) OF CHOICE: *cell cultures; culturing promoter + ~~BB8~~ promoter clones*

GEL PICTURE / OTHER SPACE:

-Mini-prep

- cell culturing

EXPECTED RESULT:

- pink cells

☐ check if APE file exists on Drive)

TEMPLATE(S): *BBa_J23100-02;*

OLIGO(S):

*RH1 RH1 RH1 RH1
PH1 PH2 PL1 PL2 PL1 PL2 PL1 PL2 PL1*

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

Name: *Coleen Tan*

CONFIRMATION?

Date: *8/8/14*

WEEKLY ONE SENTENCER:

working on APS + promote + nothing

sequencing
GEL LANE ORDER:

DAILY ONE SENTENCER:

mini prep promote + APS + nothing culture

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

mini prep

EXPECTED RESULT:

pink colonies

☐ check if APE file exists on Drive)

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

Name: *Nadu.C*

Date: *8/8/14*

CONFIRMATION?

WEEKLY ONE SENTENCER:

- Continue cloning ~~psb~~ into genes into psb1c3

GEL LANE ORDER:

DAILY ONE SENTENCER:

- Digest & ligate all genes, transform all genes into

METHOD(S) OF CHOICE:

- psb1c3*
- Digestion*
- Ligation*
- Transformation*

GEL PICTURE / OTHER SPACE:

EXPECTED RESULT:

- Transformed colonies of all genes.

☒ (check if APE file exists on Drive)

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

Name: *Ng*

CONFIRMATION?

Date: *8/12/2014*

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

Ligate: GEN Anderson Promoters with our RBS mcherry

DAILY ONE SENTENCER:

"

"

"

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

Digest Rht2 mch E⁺X

Digest Bba_12310N E⁺S

Gel extract Rht2 mch Backbone + Bba_12310N inserts

EXPECTED RESULT:

*Ligate inserts w/ Backbone
transform and plate on Cm plates*

Pink colonies tomorrow

(☐ check if APE file exists on Drive)

TEMPLATE(S): *Rht2/RL mch, Bba_12310C, Bba_12310I, Bba_12310A*

OLIGO(S): *E, X, S*

THERMOCYCLER SETTINGS: *Digest*

CONTROLS: *None*

POSITIVE: *None*

NEGATIVE: *Control Cm Plate*

*inserts were too small to bind to the spin column,
no DNA was retained for ligations*

Name: Troy

CONFIRMATION?

Date: 8/13/2014

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

Ligate iGEM Anderson Promoters with our ABS mCherry.

DAILY ONE SENTENCER:

"

"

"

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

Digest Afl2 mch Xcp

Digest Bba-J2310N SFP

Gel extract Afl2 mch insert + Bba-J2310N Backbones

EXPECTED RESULT:

Ligate insert w/ Backbones

Transform + Plate on Amp plates

Pink colonies tomorrow
(☐ check if APE file exists on Drive)

TEMPLATE(S): Afl2 mCherry, Bba-J2310C, Bba-J2310J, Bba-J2310A

OLIGO(S): X, S, P

THERMOCYCLER SETTINGS:

Digest

CONTROLS: None

POSITIVE: None

Ligate: Colonies did not grow on Antibiotic plates

NEGATIVE: Control Amp plate

Name: *Troy*

CONFIRMATION?

Date: *8/15/2014*

WEEKLY ONE SENTENCER:

GEL LANE ORDER:

Ligate existing 1GEN promoters with our ABS machinery.

DAILY ONE SENTENCER:

Resupply us with Buffers, plates, autoclaved materials.

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

Recipes

EXPECTED RESULT:

*PB Buffer Autoclaved tips + tubes
PE Buffer Clean fridge + freezer boxes
on plates*

*gel extraction
from before came
out poorly.
Continue next week!*

☐ check if APE file exists on Drive)

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

Name: *Coleman Toman*

Date: *8/15/14*

WEEKLY ONE SENTENCER:

working on prom. + RBS1 mRNA

DAILY ONE SENTENCER:

working on S/MMA gene; cloning into pS81C3

METHOD(S) OF CHOICE:

- PCR

- digestion

B.1.8

EXPECTED RESULT:

α: 1000

γ: 550

β: 1200

B: 450

(☐ check if APE file exists on Drive)

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

No template

CONFIRMATION?

gel

GEL LANE ORDER:

1 2 3 4 5 6

GEL PICTURE / OTHER SPACE:

78°C - 30" { 98°C - 10" ; 68°C - 20" ; 72°C - 2' ; 72°C - 2' ; 4°C

Name: Anirudh Joshi

CONFIRMATION?

Date: 8/15/14

WEEKLY ONE SENTENCER:

Inserting $\alpha, \beta, \delta, \beta, \gamma$ genes into

GEL LANE ORDER:

DAILY ONE SENTENCER:

pSB-1c3 backbone

~~Prod~~ Making Thin and Thick gels

GEL PICTURE / OTHER SPACE:

METHOD(S) OF CHOICE:

Gel Making * β, γ into

EXPECTED RESULT:

Gel

pSB1C3

for next week
and grow cultures
of α, β, γ

(☐ check if APE file exists on Drive)

TEMPLATE(S):

OLIGO(S):

THERMOCYCLER SETTINGS:

CONTROLS:

POSITIVE:

NEGATIVE:

CONFIRMATION?

GEL LANE ORDER:

GEL LANE ORDER:

GEL PICTURE / OTHER SPACE:

* ~~write~~ β, γ into SBIC3
Put

(☐ check if APE file exists on Drive)

THERMOCYCLER SETTINGS:

CONTROLS:

NEGATIVE:

Name: Coleen Travis

Date: 8/19/14

WEEKLY ONE SENTENCE:

working on promote + EBS + mCherry

DAILY ONE SENTENCE:

cloning β + α into pSB1c3 vector

METHOD(S) OF CHOICE:

digest E + P, ligate,

EXPECTED RESULT:

transformation

α - 1650 bp

β - 1200 bp

mCherry backbone - 2 kb.

(☐ check if APE file exists on Drive)

TEMPLATE(S):

α , β , mCherry

OLIGO(S):

THERMOCYCLER SETTINGS:

37°C - 10' ; 80°C - 20' , 4°C

CONTROLS:

POSITIVE:

NEGATIVE:

CONFIRMATION?

colonies on plates

GEL LANE ORDER:

GEL PICTURE / OTHER SPACE: