Lab note 0916-0920

0916

1. Electrophoresis

1 OneSTEP Ladder Marker 100

2 Lac

3 Lac

2. Colony PCR

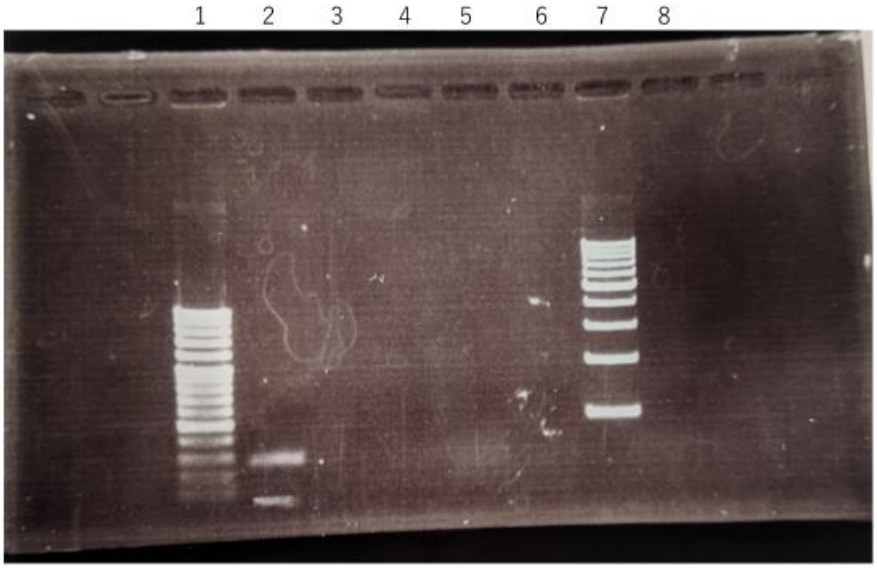
one sample

materials	concentration	volume(μL)
KOD One Pol	1U/50 μL	25
forward primer	10 μM	1.5
reverse primer	10 μM	1.5
D.W	-	up to 50
total	-	50

Cycling condition

reaction		temp.(°C)	time
cycle 1 (X1)	step 1	94	2:00
cycle 2 (X35)	step 1	98	0:10
	step 2	55	0:05
	step 3	68	0:15
cycle 3 (X1)	step 1	4	∞

Electrophoresis



1 OneSTEP Ladder Marker 100

2 Lac

3 Lac

4 pSB1C3C

5 pSB1C3C

6 pSB1C3C

7 OneSTEP Ladder Maeker 500

8 Lac

0917

1. Colony PCR

Reagent

one sample

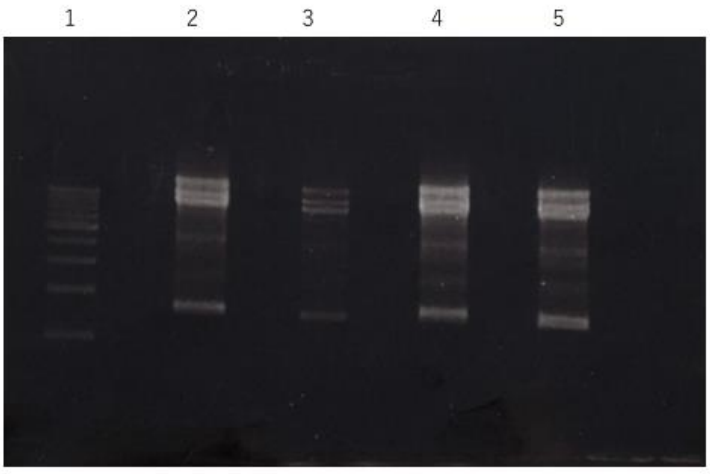
materials	volume (μL)
D.W.	22
KOD One	25
forward primer	1.5
reverse primer	1.5
total	50

Sample	forward primer	reverse primer	colony
1	Ⅶ	X	*D.radiodurans*
2	Ⅶ	X	transformant colony 3
3	Ⅶ	g	transformant colony 3

Cycling condition

reaction		temp.(°C)	time
cycle 1 (X1)	step 1	94	2:00
cycle 2 (X35)	step 1	98	0:10
	step 2	68	0:05
cycle 3 (X1)	step 1	4	∞

Electrophoresis



1 OneATEP Ladder Marker 500

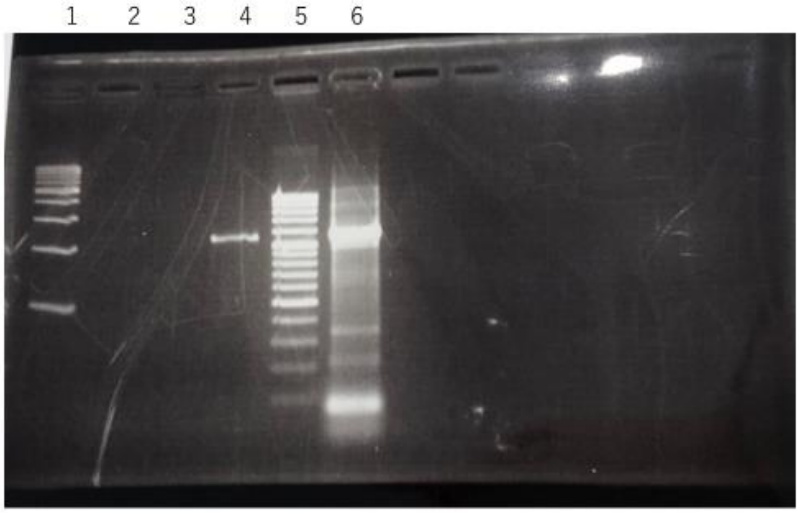
2 pSB1C3

3 pSB1C3

4 pSB1C3

0918

1. Electrophoresis



1 OneSTEP Ladder Marker 500

2 Lac+recA

3 recA

4 Lac+recA

5 OneSTEP Ladder Marker 100

6 recA

2. Gel extraction

Electrophoresis

1 OneSTEP Ladder Marker 100

2 recA

3 recA

3. PCR

one sample

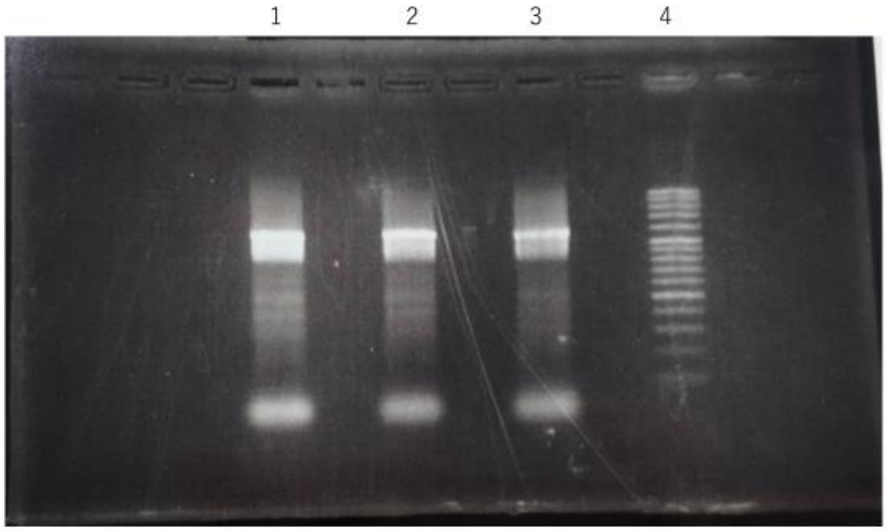
materials	concentration	volume(μL)
KOD One Pol	1U/50μL	10.0
template DNA	-	17.0
forward primer(e)	-	1.5
reverse primer(tsu)	-	1.5
D.W	-	20
total	-	50

Cycling condition

reaction		temp.(°C)	time
cycle 1 (×1)	step 1	94	2:00
cycle 2 (×35)	step 1	98	0:10
	step 2	61	0:05
	step 3	68	0:06
cycle 3 (×1)	step 1	4	∞

0919

1. Electrophoresis



1 recA

2 recA

3 recA

4 OneSTEP Ladder Marker

2. Gel extraction

1 OneSTEP Ladder Marker 100

2 recA

3 recA

4 recA

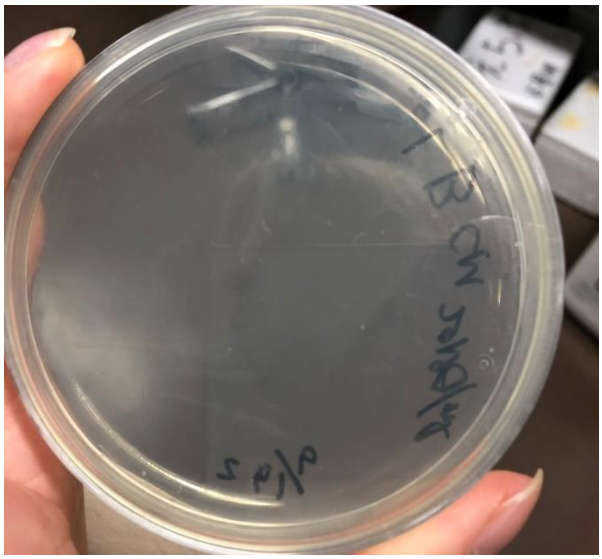
5 recA

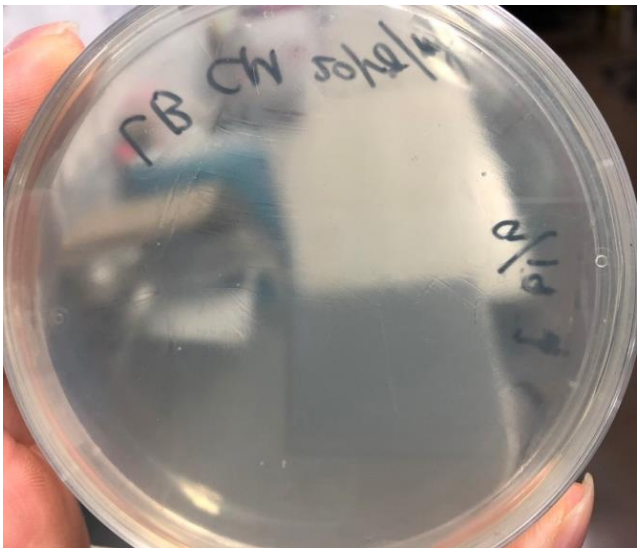
3. Infusion cloning

one sample

materials	concentration	volume(μL)
insert DNA	100 ng	5
vector DNA	100 ng	3
HD Enzyme	1X	2
dH_2O	-	up to 10
total		10

4. Transformation





0920

1. Colony PCR

Reagent

one sample

materials	concentration	volume(μL)
2×PCR buffer	1X	25
dNTP	0.4mM	10
KOD Fx Pol	1U/50μL	1.0
forward primer(h)	0.15~0.3μM	1.5
reverse primer(g)	0.15~0.3μM	1.5
D.W	-	up to 50
total	-	50

Cycling condition

reaction		temp.(°C)	time
cycle 1 (X1)	step 1	94	2:00
cycle 2 (X35)	step 1	98	0:10
	step 2	68	1:00
cycle 3 (X1)	step 1	4	∞