PCR protocol

Sheet ID PS_05282
Release date : 3/3/2014
Print date : 3/3/2014

RBRC 04878 Strain name B6;129-Tg(CMV-EGFP,U6-shNanog)1Ttada

Primer	primer_name	primer_seq	length		
1	pSico_F	CGCACAGACTTGTGGGAGAAG	21		Depositor's primer
2	PSico_R	CAAACACAGTGCACACCACGC	21		Depositor's primer
3	mLC3ex4AG	GTTAGCATTGAGCTGCAAGCGCCGTCT	27		Mol. Biol. Cell., 15, 1101-1111(2004).
4	mLC3ex3GT	TGAGCGAGCTCATCAAGATAATCAGGT	27		Mol. Biol. Cell., 15, 1101-1111(2004).

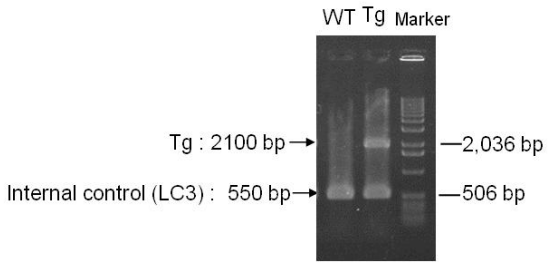
Reaction Components

Enzyme: TOYOBO KOD FX (Code.No : KFX-101)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		0.8			
PCR buffer	2 X	10.0			
dNTP	2.0 mM	4.0			
primer 1	10 pmol/μl	1.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
primer 4	10 pmol/μl	1.0			
DNA		1.0			
Taq	1 U/μl	0.2			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10	*
3	68	180	* *Repeat for 30 cycles



Marker

1 kb DNA ladder (Invitrogen, Cat. 15615-024)

Gel

1% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 1	primer 2	2,100 kb	Tg
primer 3	primer 4	550 bp	Internal control (LC3)

Comment Novagen KOD Xtreme™ Hot Start DNA Polymerase can be purchased from EMD.

PCR protocol

Sheet ID PS_05027
Release date : 12/20/2012
Print date : 3/3/2014

RBRC 04878 Strain name B6;129-Tg(CMV-EGFP,U6-shNanog)1Ttada

Primer	primer_name	primer_seq	length		
1	pSico_F	CGCACAGACTTGTGGGAGAAG	21		Depositor's primer
2	PSico_R	CAAACACAGTGCACACCACGC	21		Depositor's primer
3	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24		JAX Genotyping Protocol
4	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25		JAX Genotyping Protocol

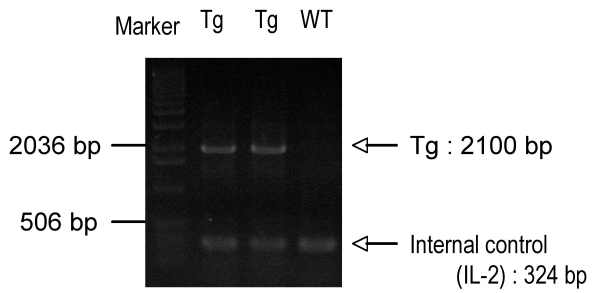
Reaction Components

Enzyme: TaKaRa LA Taq with GC buffer (Cat. No. RR02AG)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.9			
PCR buffer	2 X GC I	10.0			
dNTP	2.5 mM	2.0			
primer 1	10 pmol/μl	1.5			
primer 2	10 pmol/μl	1.5			
primer 3	10 pmol/μl	0.5			
primer 4	10 pmol/μl	0.5			
DNA		1.0			
Taq	5U/μl	0.1			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30	*
3	60	30	*
4	72	120	* *Repeat for 30 cycles
5	72	300	



Marker: 1 kb DNA ladder (Invitrogen, Cat. 15615-024)

Gel: 1% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 1	primer 2	2100 bp	Tg
primer 3	primer 4	324 bp	Internal control (IL-2)

Comment

PCR protocol

Sheet ID PS_04064
Release date : 12.02.2011
Print date : 3/3/2014

RBRC 04878 Strain name B6;129-Tg(CMV-EGFP,U6-shNanog)1Ttada

Primer	primer_name	primer_seq	length		
1	pSico_F	CGCACAGACTTGTGGGAGAAG	21		Depositor's primer
2	PSico_R	CAAACACAGTGCACACCACGC	21		Depositor's primer
3	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24		JAX Genotyping Protocol
4	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25		JAX Genotyping Protocol

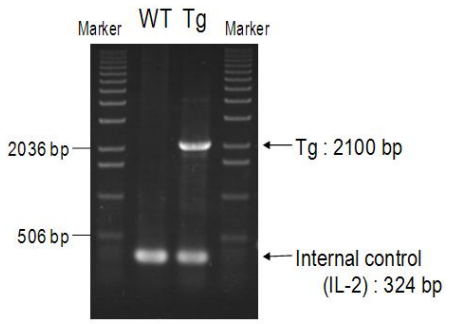
Reaction Components

Enzyme: TaKaRa LA Taq with GC buffer (Cat. No. RR02AG)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.9			
PCR buffer	2 X GC I	10.0			
dNTP	2.5 mM	2.0			
primer 1	10 pmol/μl	1.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
primer 4	10 pmol/μl	1.0			
DNA		1.0			
Taq	5U/μl	0.1			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30	*
3	60	30	*
4	72	120	* *Repeat for 30 cycles
5	72	300	



Marker

1 kb DNA ladder (Invitrogen, Cat. 15615-024)

Gel

1% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 1	primer 2	2100 bp	Tg
primer 3	primer 4	324 bp	Internal control (IL-2)

Comment