PCR protocol

Sheet ID PS_04812
Release date : 7/25/2014
Print date : 7/25/2014

RBRC 02220 Strain name B6;129-Mafb<tm(GFP)UTR>

Primer	primer_name	primer_seq	length		
1	hn2	AGCAGCCGATTGTCTGTTGTGCCCAGTCAT	30		Depositor's primer
2	mafB cds-as	CCATCCAGTACAGGTCCTCG	20		Depositor's primer
3	MAFB cds-s	TGAGCATGGGGCAAGAGCTG	20		Depositor's primer

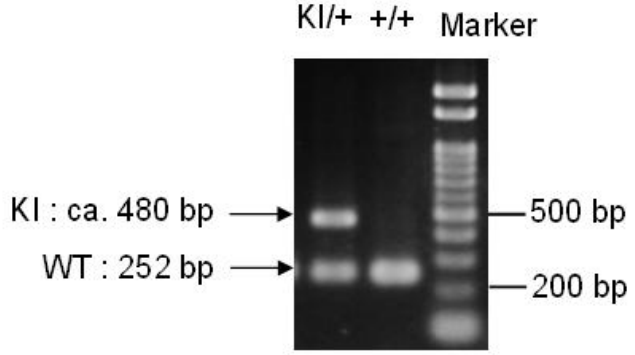
Reaction Components

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

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		1.8			
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			
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	65	30	*
4	68	30	* *Repeat for 35 cycles



Marker

Gene Ladder 100 (Wako, Cat. 316-06951)

Gel

2% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 2	primer 3	252 bp	WT
primer 2	primer 1	ca. 480 bp	KI

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

PCR protocol

Sheet ID PS_01272
Release date : 3/23/2012
Print date : 7/25/2014

RBRC 02220 Strain name B6;129-Mafb<tm(GFP)UTR>

Primer	primer_name	primer_seq	length		
1	hn2	AGCAGCCGATTGTCTGTTGTGCCCAGTCAT	30		Depositor's primer
2	mafB cds-as	CCATCCAGTACAGGTCCTCG	20		Depositor's primer
3	MAFB cds-s	TGAGCATGGGGCAAGAGCTG	20		Depositor's primer

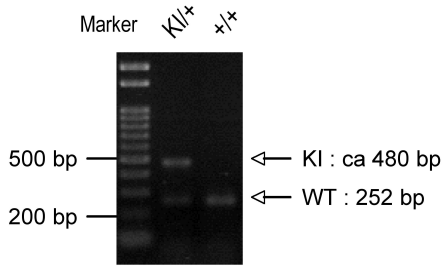
Reaction Components

Enzyme: Roche Taq (Cat. No. BM1418432)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		12.3			
PCR buffer	10 X	2.0			
dNTP	2.5 mM	1.6			
primer 1	10 pmol/μl	1.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
DNA		1.0			
Taq	5 U/μl	0.1			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30	*
3	65	30	*
4	72	60	* *Repeat for 35 cycles
5	72	300	



Marker

Gene Ladder 100 (Wako, Cat. 316-06951)

Gel

2% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 2	primer 3	252 bp	WT
primer 2	primer 1	ca 480 bp	KI

Comment