PCR protocol

Sheet ID PS_07215
Release date : 10/5/2017
Print date : 10/5/2017

RBRC 09592 **Strain name** B6-ROSA26Tm(SAEiCE)13Card

Primer	primer_name	primer_seq	length		
1	R26F2	AAAGTCGCTCTGAGTTGTTAT	21		Nature Genetics 21, 70-71 (1999).
2	olMR0316	GGAGCGGGAGAAATGGATATG	21		JAX genotyping protocol/Nature Genetics
3	iCre-2: 5'	TTGGTCCTGCCAATGTGGATC	21		Depositor's primer
4	iCre-1: 5'	AGGCCTTCTCTGAACACACC	20		Depositor's primer

Reaction Components

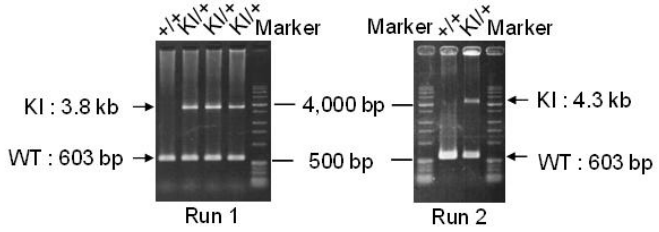
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		1.8	1.8		
PCR buffer	2 X	10.0	10.0		
dNTP	2.0 mM	4.0	4.0		
primer 1	10 pmol/μl	1.0	1.0		
primer 2	10 pmol/μl	1.0	1.0		
primer 3	10 pmol/μl	1.0	0		
primer 4	10 pmol/μl	0	1.0		
DNA		1.0	1.0		
Taq	1 U/μl	0.2	0.2		
	total	20	20		

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10	*
3	60	30	*
4	68	300	* *Repeat for 30 cycles

Standard PCR



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	603 bp	WT
primer 1	primer 3	3.8 kb	KI
primer 2	primer 4	4.3 kb	KI

Comment

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