PCR protocol

Sheet ID PS_05396
Release date : 2/6/2014
Print date : 2/6/2014

RBRC 01929 **Strain name** B6;129-Rab8b<tm1>

Primer	primer_name	primer_seq	length		
1	8bWT-S1	CAGAAGCCCTTCCTCGTTCCTGG	23		Depositor's primer
2		CCGTCGAGTTCTATCGTTCTG	21		RIKEN BRC
3	NP4	TGCTCGACGTTGTCACTGAAGC	22		Depositor's primer

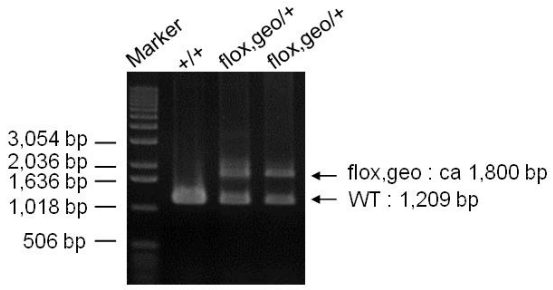
Reaction Components

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	60	30	*	
4	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	1,209 bp	WT
primer 2	primer 3	ca 1,800 bp	flox,geo

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