PCR protocol

RBRC 03481 **Strain name** B6.129P2-Chek2<tm1Nmt>

Primer

	primer_name	primer_seq	length		
1		CTCGCTGACCTAGGTAGCAGGACC	24		Depositor's primer
2		TGTGCCGGTAGAGGAGCTGG	20		Depositor's primer
3		GGGTGGGGTGGGATTAGATAAATG	24		Depositor's primer

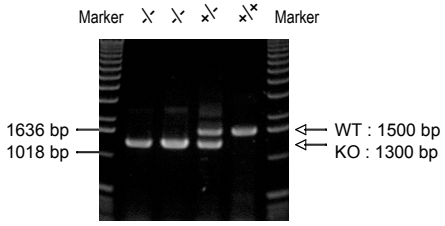
Reaction Components

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

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		3.8			
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			
DNA		1.0			
Taq	5U/μl	0.2			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30	*
3	63	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	1500 bp	WT
primer 1	primer 3	1300 bp	KO

Comment