

# PCR protocol

RBRC 04885

Strain name B6;129S4-Cacna1e<tm1Ttan>

Primer	primer_name	primer_seq	length	
1	TTB25F1	CTGTCTGTTTAAACTTCAGGATGG	24	Depositor's primer
2	TTB25R1	GGCTTAAGTGGTGGAAAAGAGA	22	Depositor's primer
3	lacZR1	GTGCGGGCCTCTTCGCTATTACGC	24	Depositor's primer

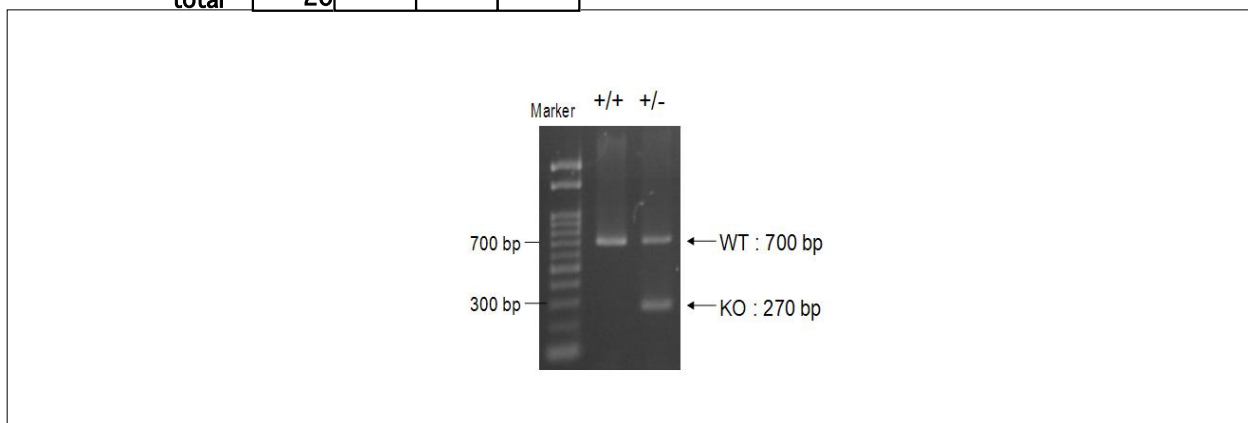
**Reaction Components**

**Reaction Conditions**

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

	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/ $\mu$ l	1.0			
primer 2	10 pmol/ $\mu$ l	1.0			
primer 3	10 pmol/ $\mu$ l	1.0			
DNA		1.0			
Taq		0.1			
total		20			

step	temp (°C)	time (sec)	note
1	94	180	
2	94	30*	
3	60	30*	
4	72	60*	*Repeat for 30 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 1	primer 2	700 bp	WT
primer 1	primer 3	270 bp	KO

Comment