

# PCR protocol

RBRC 02178

Strain name STOCK Cd52&lt;tm10sb&gt;

Primer	primer_name	primer_seq	length	
1	02178_WT	CTTGCCCAGTGATAGAAGGCATGTGGTC	28	RIKEN BRC
2	PrimerA	TGAATTCTTCAAAGTGGCCTGCAGACTGTC	30	Depositor's primer
3	Neo_R	GTCCTGATAGCGGTCCGCCACAC	23	RIKEN BRC

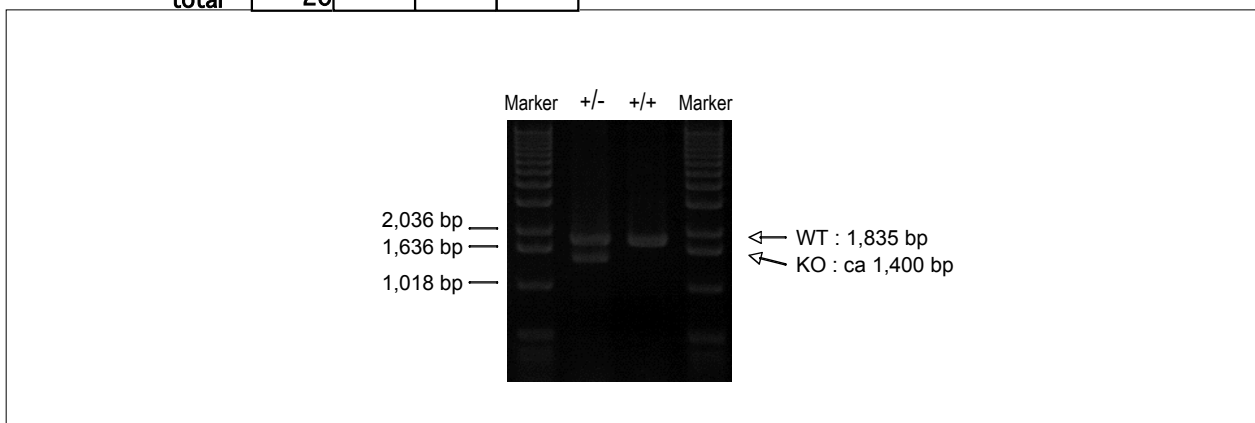
## Reaction Components

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

## Reaction Conditions

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		3.9			
PCR buffer	2 X GC I	10.0			
dNTP	2.5 mM	2.0			
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	5U/ $\mu$ l	0.1			
<b>total</b>		<b>20</b>			

step	temp (°C)	time (sec)	note
1	94	180	
2	94	30*	
3	60	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	1,835 bp	WT
primer 1	ca 1400 bp	KO

Comment