

RBRC 01047

Strain name B6.Cg-Creb5<tm1Sis>/Rbrc

Primer	primer_name	primer_seq	length	Reference
1	BPaEX2	AGCTCAAGGTCTCTTGCTCTCACAAGTGTG	30	Depositor's primer
2	intron 9	GATGCTGGATGAGAGGGCCTGG	22	RIKEN BRC
3	Neo30	TTCTATCGCCTTCTTGACGAGTTCTTCTGA	30	Depositor's primer

Reaction Components

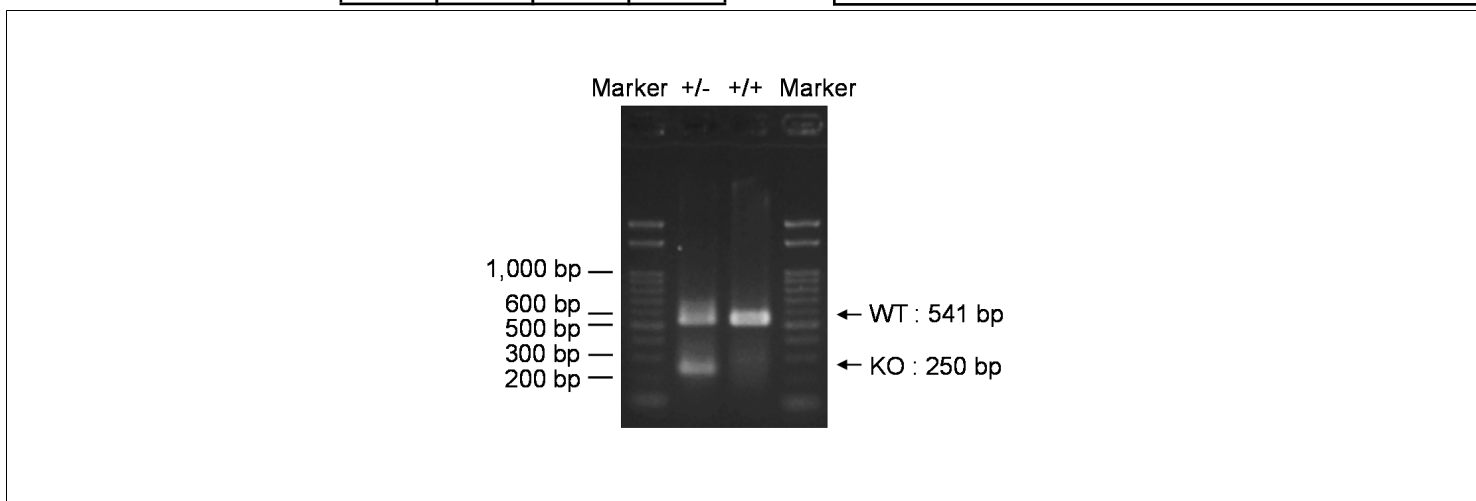
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

Reagent	Conc	Reaction 1	2	3	4	(μ L)
H2O		1.8				
PCR buffer	2 X	10.0				
dNTP	2.0 mM	4.0				
primer 1	10 pmol/ μ l	1.0				
primer 2	10 pmol/ μ l	1.0				
primer 3	10 pmol/ μ l	1.0				
DNA		1.0				
Taq	1 U/ μ l	0.2				
total		20				

Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	65	30*	
4	68	60*	*Repeat for 30 cycles

Marker Gene Ladder 100 (Wako, Cat. 316-06951)
 Gel 2% agarose in 1 X TAE (100 V, 30 min)



primer_set	product_size	product (allele type)	
primer 1	primer 2	541 bp	WT
primer 1	primer 3	250 bp	KO

+/+ (wild-type): 541 bp
 +/- (heterozygous): 541 bp and 250 bp

Comment [Standard PCR](#)

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