

RBRC 01868

Strain name Pgds-Cre

Primer	primer_name	primer_seq	length	Reference
1	exon1_F	CATCAGGCCCCAGGCACCTGC	20	RIKEN BRC
2	intron1_R	CAGGCTGACAGTGCCCGAGAAC	21	RIKEN BRC
3	Cre2	CTACACCAGAGACGGAAATC	20	Depositor's primer

Reaction Components

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

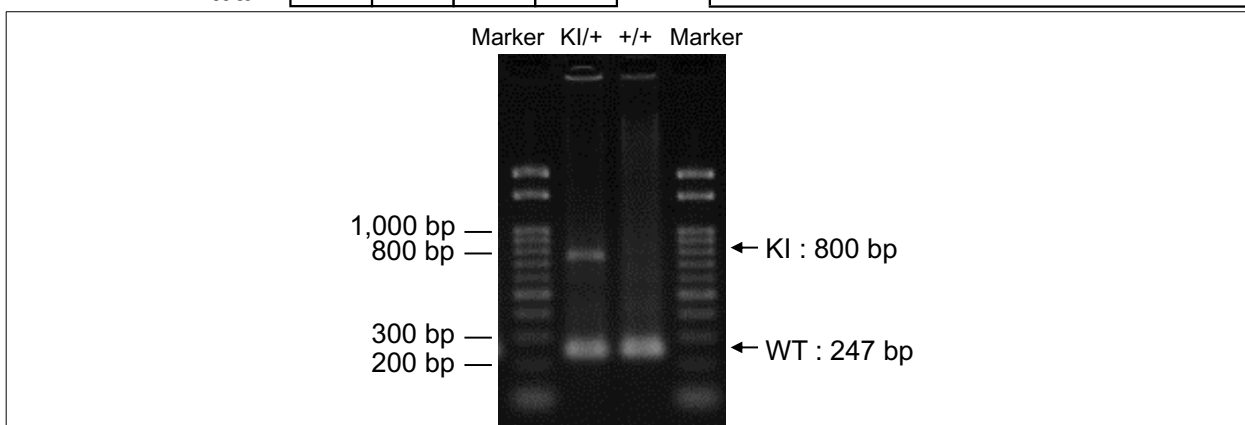
Reaction Conditions

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

Reagent	Conc	Reaction 1	2	3	4
H ₂ O		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			

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	247 bp	WT	+/+ : 247 bp KI/+ : 247 bp, 800 bp
primer 1	800 bp	KI	

Comment [Standard PCR](#)

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

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Primer	primer_name	primer_seq	length	Reference
1	Cre1	AATGCTTCTGTCCGTTTGCC	20	Depositor's primer
2	Cre2	CTACACCAGAGACGGAAATC	20	Depositor's primer
3	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24	Internal control (IL-2) JAX Genotyping Protocol
4	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25	Internal control (IL-2) JAX Genotyping Protocol

Reaction Components

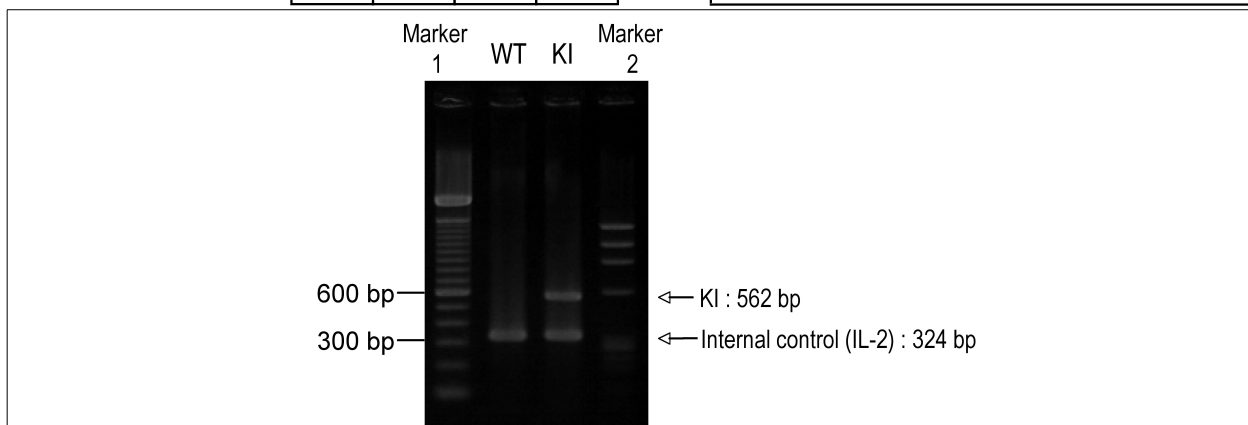
Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	180	
2	94	30*	
3	60	30*	
4	72	60*	*Repeat for 35 cycles
5	72	300	

Reagent	Conc	Reaction 1	2	3	4
H2O		15.3			
PCR buffer	10 X	2.5			
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			
primer 4	10 pmol/μl	1.0			
DNA		1.0			
Taq		0.2			
total		25			

Marker marker 1:100 bp ladder (Invitrogen, Cat. 15628-050), marker2: φX174 Hae III
Gel 2% agarose in 1 X TAE (100 V, 35 min)



primer_set	product_size	product (allele type)
primer 1	562 bp	KI
primer 3	324 bp	Internal control (IL-2)

Comment Standard PCR