

RBRC 11579

Strain name C57BL/6N-Gt(ROSA)26Sor<tm1.1(CAG-cas9)Mabe>

Primer	primer_name	primer_seq	length	Reference
1	ROSA26 B1F2	AGTCGCTCTGAGTTGTTATC	20	Depositor's primer
2	ROSA26 B2R1	GGATCTCAAGCAGGAGAGTA	20	Depositor's primer
3		GTCTATCGGTGTTTCCCAGGACC	23	RIKEN BRC

Reaction Components

Enzyme

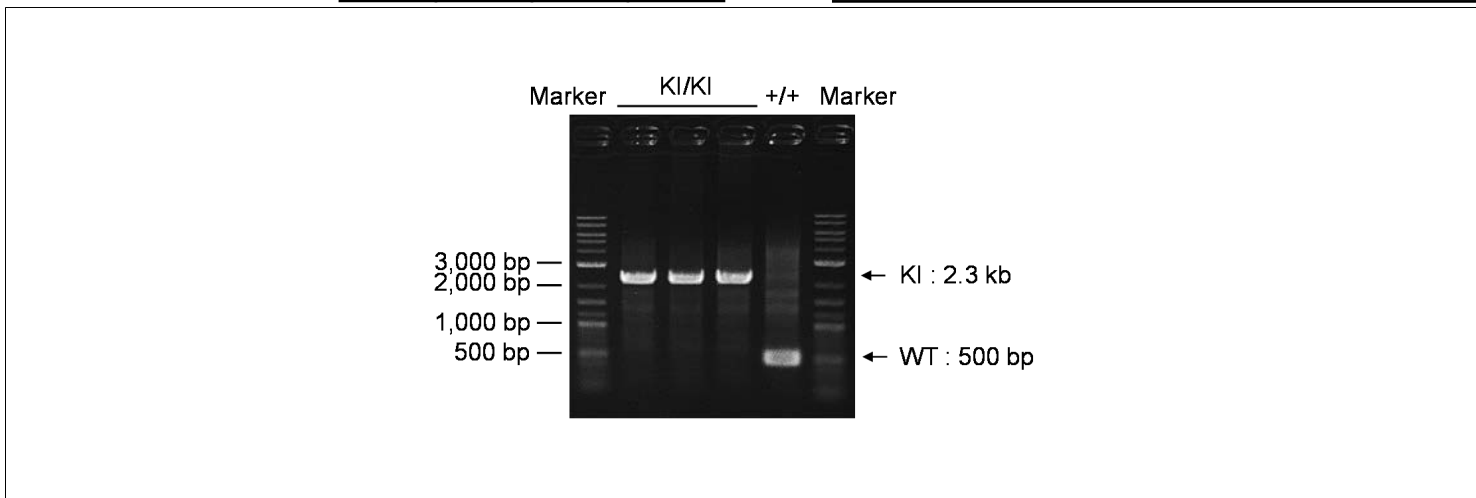
Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30	
4	68	180*	*Repeat for 30 cycles

Reagent	Conc	Reaction 1	2	3	4	(μL)
H2O		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

Gel



primer_set	product_size	product (allele type)	
primer 1	primer 2	500 bp	WT
primer 1	primer 3	2.3 kb	KI

+/+ (wild-type): 500 bp
KI/KI (homozygous): 2.3 kb

Comment

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