

RBRC 06257

Strain name C57BL/6-Gt(ROSA)26Sor<tm1(H4K20me1-mCherry)Osb>/44

Primer	primer_name	primer_seq	length	Reference
1	ROSA26-NotI-Fw	GAGCGGCCGCCACCCCTCCCCTTCTCTGG	30	Depositor's primer
2	ROSA26-NruI-Rev	CCTCGCGACACTGTATTTTCATACTGTAGTA	30	Depositor's primer
3	mCherry-300bp-rv	TTCAGCCTCTGCTTGATCTCGCCCTTC	27	Depositor's primer

**Reaction Components**

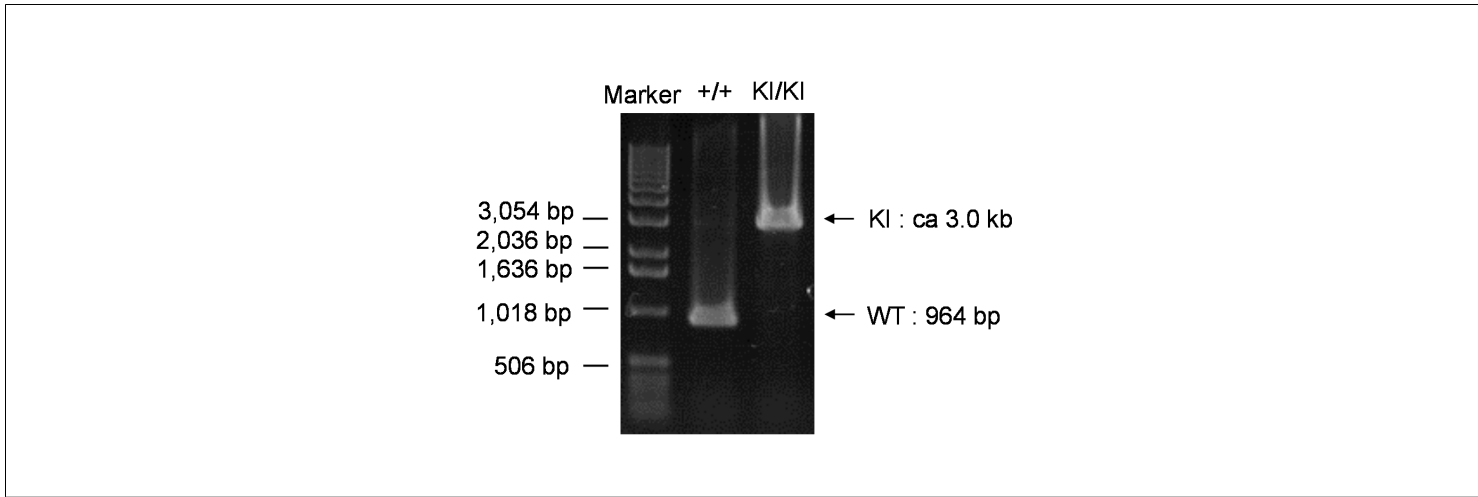
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

Reagent	Conc	Reaction 1	2	3	4	(μL)
H2O		1.6				
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	0.4				
<b>total</b>		20				

**Reaction Conditions**

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30*	
4	68	300*	*Repeat for 30 cycles

**Marker** 1 kb DNA ladder (Invitrogen, Cat. 15615-024)  
**Gel** 1% agarose in 1 X TAE (100 V, 30 min)



primer_set	product_size	product (allele type)
primer 1 / primer 2	964 bp	WT
primer 1 / primer 3	ca 3.0 kb	KI

**Comment** Standard PCR

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