PCR protocol

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RBRC 09688 Strain name C.Cg-Siglech<tm1.1Ksat>

Primer	primer_name	primer_seq	length		
1	Siglech-exon5-FW	GATTAGTCTCTCAGGTGAGTGATATGG	27		Immunity, 35, 958-971 (2011).
2	Siglech-exon5-Rev	GACATATTTCAGTTAACAAATACAGTTCGT	30		Immunity, 35, 958-971 (2011).

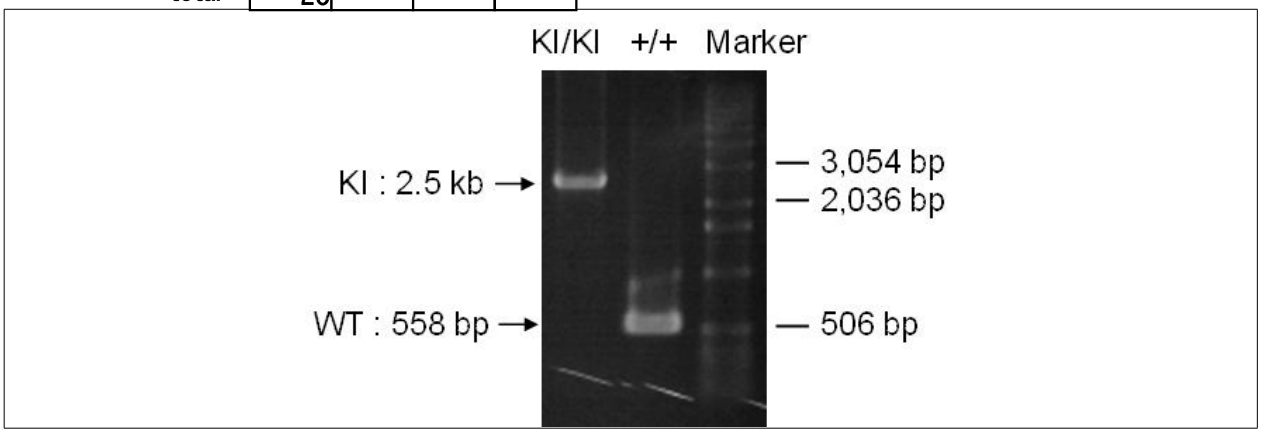
Reaction Components

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

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.8			
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			
DNA		1.0			
Taq	1 U/μl	0.2			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10	*
3	60	30	*
4	68	240	* *Repeat for 30 cycles



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	primer 2	558 bp	WT
primer 1	primer 2	2.5 kb	KI

Comment