PCR protocol

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RBRC 04197 **Strain name** B6.129P2-Cxcr4<tm1Tng>

Primer	primer_name	primer_seq	length		Reference
1	CXCR4 Foward	AGACAGACGTGAGAAAAACAAGGTT	25		Depositor's primer
2	CXCR4 Wild Reverse	TGATGAAGTAGATGGTGGGCAGGAA	25		Depositor's primer
3	Neo	GCAATCCATCTTGTTCAATGGCC	23		Depositor's primer

Reaction Components

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

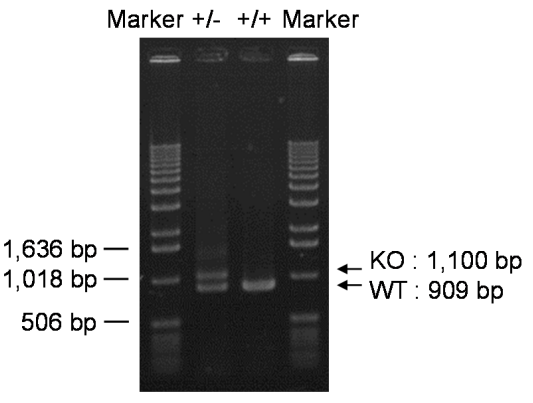
Reagent	Conc	Reaction 1	2	3	4	
H2O		12.3				(μL)
PCR buffer	10 X	2.0				
dNTP	2.5 mM	1.6				
primer 1	10 pmol/μl	1.0				
primer 2	10 pmol/μl	1.0				
primer 3	10 pmol/μl	1.0				
DNA		1.0				
Taq	5 U/μl	0.1				
	total	20				

Reaction Conditions

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

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	909 bp	WT	+/+ (wild-type): 909 bp +/- (heterozygous): 909 bp and 1,100 bp
primer 1	primer 3	1,100 bp	KO	

Comment Standard PCR