PCR protocol

RBRC 02327 **Strain name** NOD.129P2(B6)-Fcer1g<tm1Rav>

Primer

	primer_name	primer_seq	length	
1	Neo	CTCGTGCTTTACGGTATCGCC	21	
2	5'EC1(F)	CCTACTCTACTGTCGACTCAAG	22	
3	3'EC1(R)	GGCTGGCTATAGCTGCCTTTC	21	

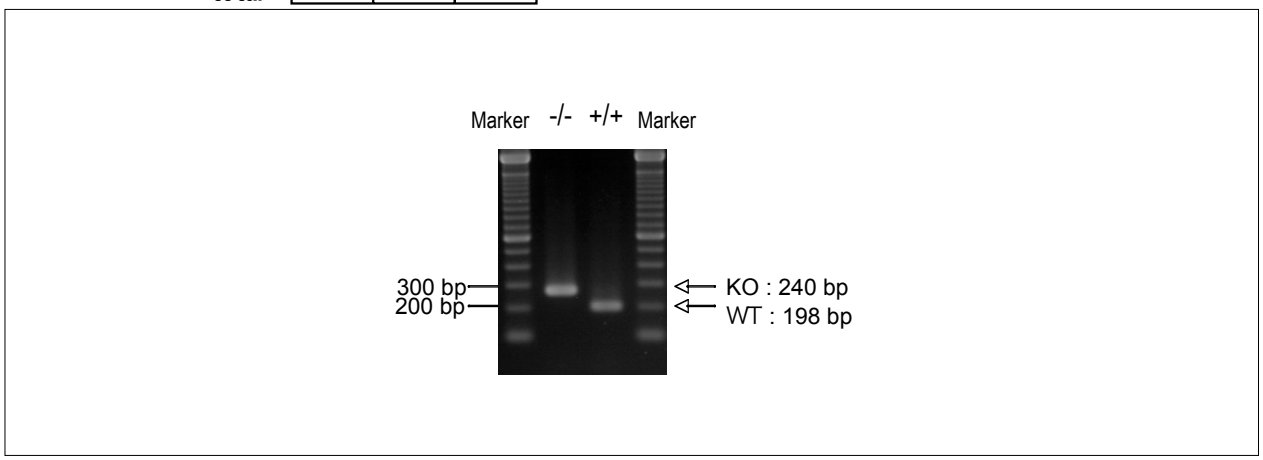
Reaction Components

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

	Conc	Run 1	Run 2	Run 3
H2O		16.3		
PCR buffer	10 X	2.5		
dNTP	2.5 mM	2.0		
primer 1	10 pmol／μ	1.0		
primer 2	10 pmol／μ	1.0		
primer 3	10 pmol／μ	1.0		
DNA		1.0		
Taq		0.2		
	total	25		

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30*	
3	63	30*	
4	72	60*	*repeat for 30 cycles
5	72	300	



Marker

100 bp ladder (Invitrogen, Cat. 15628-050)

Gel

2% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 2	primer 3	198 bp	wild
primer 1	primer 3	240 bp	KO

Comment