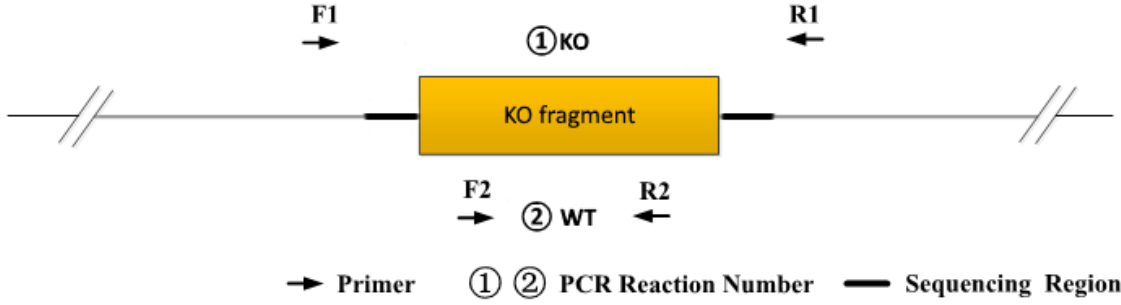


Genotyping Report

Strain ID	T030984	Strain Type	KO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Zifan Lin	Gene Name	<i>Itgad</i>		

1. Strategy of Genotyping



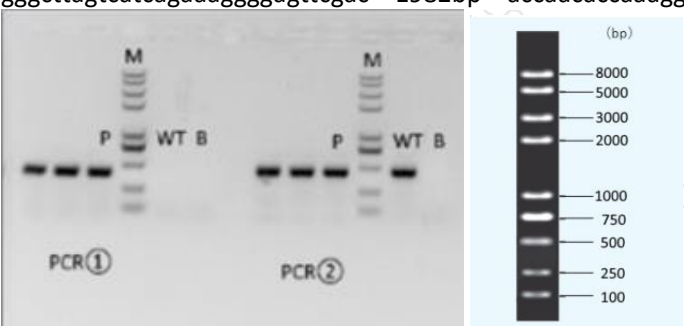
Wild type: ①PCR reaction obtains a single WT band; ②PCR reaction obtains a single WT band.
Heterozygote: ①PCR reaction obtains a WT band and a KO band; ②PCR reaction obtains a WT band.
Homozygote: ①PCR reaction obtains a single KO band; ② PCR reaction without product.
Note: 1)The sizes of WT and Targeted band are shown below.
2)If the WT band is too large, it may not be possible to obtain a WT band.

2. Primer Information

PCR No.	Primer No.	Sequence	Band Size
PCR①	T030984-F1	TAGACTCCAGAATCCAGAGACGGTTG	WT:2392bp KO: 410bp
	T030984-R1	AGGTGACTCCAGGGTTTGTGAC	
PCR②	T030984-F2	TAGTGTTCTAGTGTCAGGCTCCTAGC	WT:445bp KO:0bp
	T030984-R2	TGTGTAACCTGTGGCTGTCGATGC	

3. Gel Image

gggcttagtcatcagaaaggggagttcgac---1982bp---accaacaccaaaggtagccattttatcctc



Note: P: Positive control; WT: Wildtype control; B: Blank control (ddH₂O); M: DNA Ladder



4. PCR Condition

PCR Reaction Component			
Seg.	reaction component	Volume (μ l)	
1	2 \times Taq Master Mix , Dye Plus, (Vazyme P112-03)	12.5	
2	ddH ₂ O	9.5	
3	Primer A(10 μ mol/ μ l)	1	
4	Primer B(10 μ mol/ μ l)	1	
5	Template(\approx 100ng/ μ l)	1	
PCR program			
Seg.	Temp.	Time	Cycle
1	95 $^{\circ}$ C	5min	
2	98 $^{\circ}$ C	30s	20 \times
3	65 $^{\circ}$ C (-0.5 $^{\circ}$ C/cycle)	30s	
4	72 $^{\circ}$ C	45s	
5	98 $^{\circ}$ C	30s	
6	55 $^{\circ}$ C	30s	20 \times
7	72 $^{\circ}$ C	45s	
8	72 $^{\circ}$ C	5min	
9	10 $^{\circ}$ C	hold	

Reviewer:Ting Sun

Date:2021-6-10