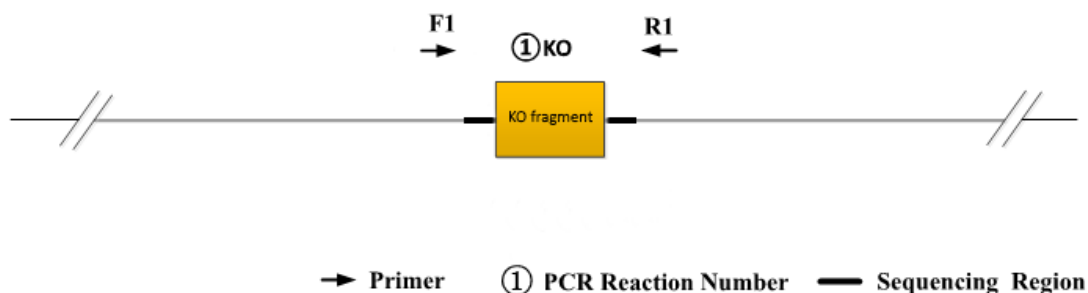


Genotyping Report

Strain ID	T001747	Strain Type	CKO(Cas9)	Genetic Background	C57BL/6J
Designer	Ying Chen	Gene Name	<i>Atg4c</i>		

1. Strategy of Genotyping



Wild type: ①PCR reaction obtains a single WT band.

Heterozygote: ①PCR reaction obtains a WT band and a KO band.

Homozygote: ①PCR reaction obtains a single KO band.

Note : 1)The sizes of WT and Targeted band are shown below.

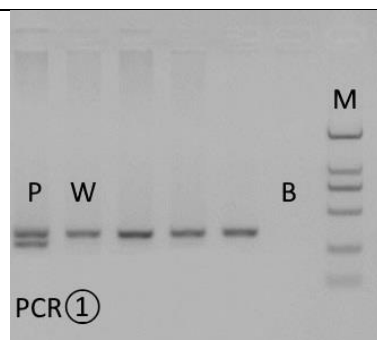
2)If the KO fragment $\leq 20\text{bp}$, the genotype should be confirmed by Sequencing.

Sequence : gagaactcagaactccagcag---56bp---gcttatcacaggaagatcatctc

2. Primer Information

PCR No.	Primer Name	Sequence	Band Size
PCR①	F1	TGCACATTGAGAACGCAGACT	WT: 341bp KO: 285bp
	R1	GCACAAGGCAGAAGAGTTGG	

3. Gel Image



Note: WT: Wildtype control; N: Blank control (ddH₂O); M:DNA Ladder

4. PCR Condition

PCR Reaction Component			
Seg.	reaction component		Volume (μl)
1	2 × Taq Master Mix , Dye Plus, (Vazyme P112-03)		12.5
2	ddH2O		9.5
3	Primer A(10pmol/μl)		1
4	Primer B(10pmol/μl)		1
5	Template(≈100ng/μl)		1
PCR program			
Seg.	Temp.	Time	Cycle
1	95℃	5min	20×
2	98℃	30s	
3	65℃ （-0.5℃/cycle）	30s	
4	72℃	45s	
5	98℃	30s	20×
6	55℃	30s	
7	72℃	45s	
8	72℃	5min	
9	10℃	hold	

Reviewer:Ting Sun

Date:2020-4-23