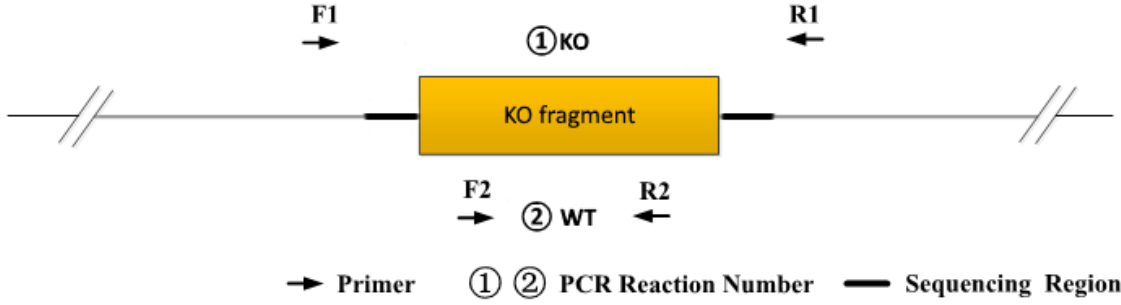


## Genotyping Report

Strain ID	T031436	Strain Type	KO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Zifan Lin	Gene Name	<i>Cry11</i>		

### 1. Strategy of Genotyping



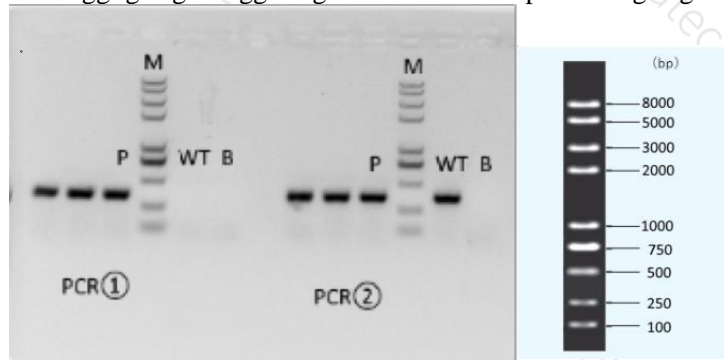
Wild type: ①PCR reaction obtains a single WT band; ②PCR reaction obtains a single WT band.  
 Heterozygote: ①PCR reaction obtains a WT band and a KO band; ②PCR reaction obtains a WT band.  
 Homozygote: ①PCR reaction obtains a single KO band; ② PCR reaction without product.  
 Note: 1)The sizes of WT 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	T031436-F1	ATGCCACGAAATCTGCTGTCTCAC	Wt:3414bp Targeted:373bp
	T031436-R1	AACAGGGTCCACACACGAAAC	
PCR②	T031436-F2	CAGGAAGGAGATGAAATCCCTG	WT:344bp Targeted:0bp
	T031436-R2	GGTCGTA CTACCTTTAATCTCAGC	

### 3. Gel Image

tcttcccggttgactgcccttggcaactgtatccta-----3041bp-----ccggtccgtcttaagtataactgtcactttgtgct



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



#### 4. PCR Condition

PCR Reaction Component			
Seg.	reaction component	Volume ( $\mu$ l)	
1	2 $\times$ Taq Master Mix , Dye Plus, (Vazyme P112-03)	12.5	
2	ddH <sub>2</sub> O	9.5	
3	Primer A(10 $\mu$ mol/ $\mu$ l)	1	
4	Primer B(10 $\mu$ mol/ $\mu$ l)	1	
5	Template( $\approx$ 100ng/ $\mu$ 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:2021-6-4