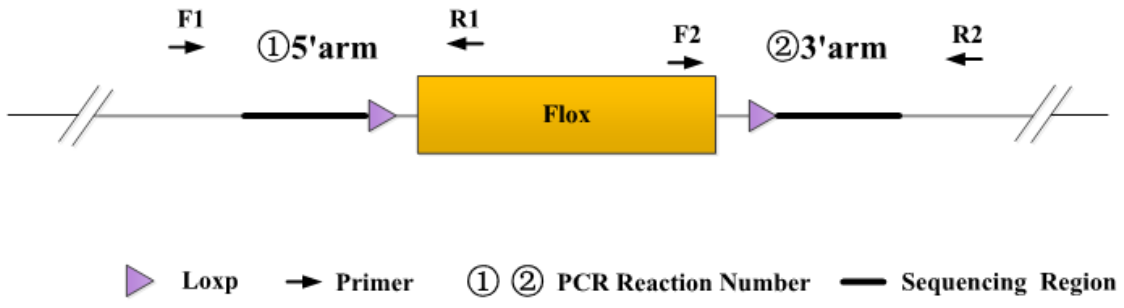
Genotyping Report

Strain ID	T012998	Strain Type	CKO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Ya'nan Xu	Gene Name	*Cfh*		

1. Strategy of Genotyping

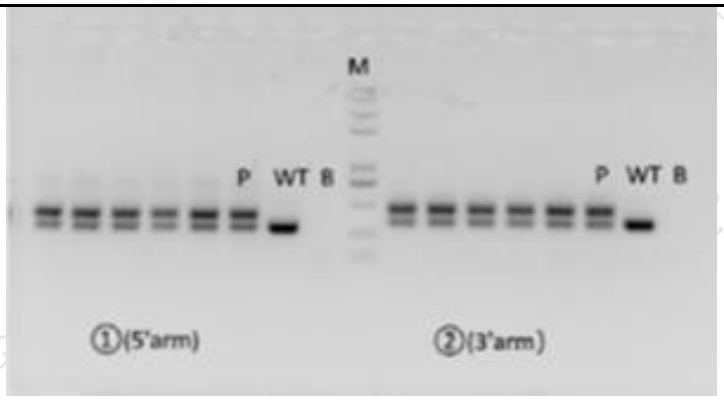


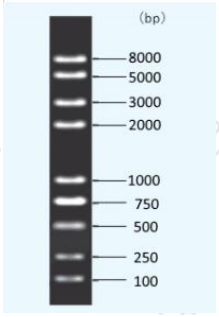
Wild type: ①PCR reaction obtains a single WT band; ②PCR reaction obtains a single WT band.
Heterozygote: ①PCR reaction obtains a WT band and a Targeted band; ②PCR reaction obtains a WT band and a Targeted band.
Homozygote: ①PCR reaction obtains a single Targeted band; ②PCR reaction obtains a single Targeted band.
Note: The sizes of WT and Targeted band are shown below.

2. Primer Information

PCR No.	Primer No.	Primer Name	Sequence	Band Size
①(5'arm)	F1	JS04346-Cfh-5wt-tF1	CCATCTGGAGTGTCATACCTTTA	WT:291bp Targeted:393bp
	R1	JS04346-Cfh-5wt-tR1	TCTTCTCCCTCGCCCATCTCT	
②(3'arm)	F2	JS04346-Cfh-3wt-tF1	TAGGGAAAAGAATTGTGAAAACA	WT:376bp Targeted:479bp
	R2	JS04346-Cfh-3wt-tR1	TTTTTGGAGCTAATTCTAAAGTCTG	

3. Gel Image & Conclusion





Note: P: Positive control; WT: Wildype control; B: Blank control (ddH_2O); 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	
2	98℃	30s	20×
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:2020-5-12