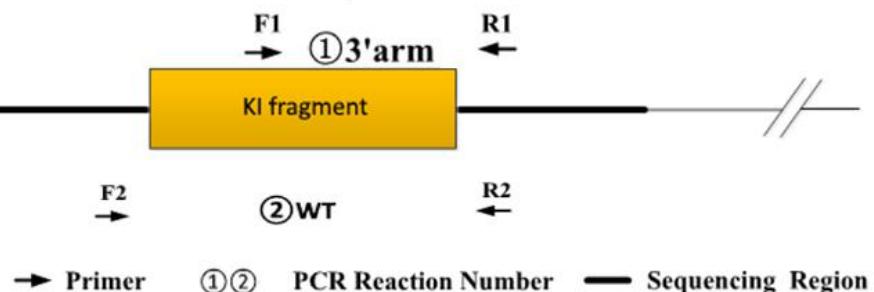




Genotyping Report

Strain ID	T004833	Strain Type	KI(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Dongdong Zhang	Gene Name			<i>hKRT14-iCre-ployA</i>

1. Strategy of Genotyping



Wild type: ①PCR reaction obtains none band; ②PCR reaction obtains a WT band.

Heterozygote: ①PCR reaction obtains a Targeted band; ②PCR reaction obtains a WT band.

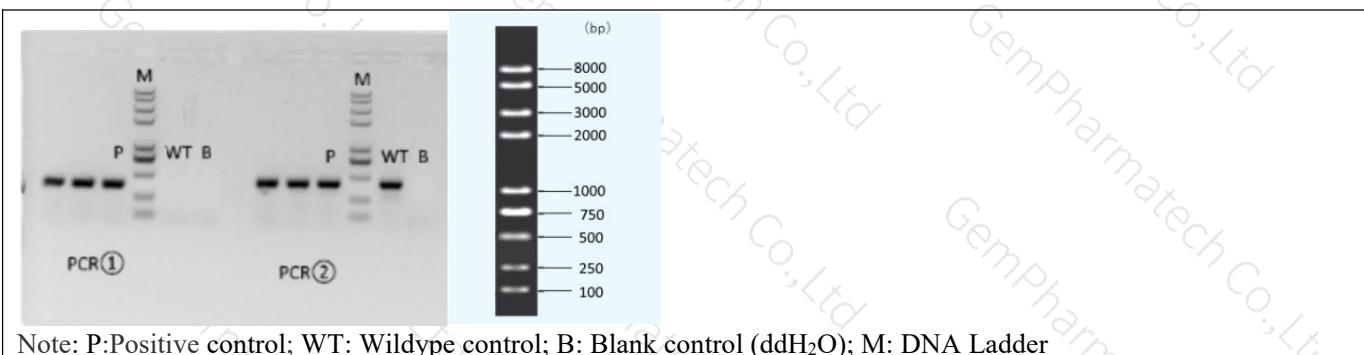
Homozygote: ①PCR reaction obtains a Targeted band; ②PCR reaction obtains none band.

Note: The sizes of WT and Targeted band are shown below. For ②PCR reaction, because the WT band is much smaller than the target band, it is likely to produce dominant amplification, the reaction is only used to judge whether there is a WT allele.

2. Primer Information

PCR No.	Primer No.	Sequence	Band Size
①3'arm	T004833-F1	TTACCGGAGGTCCATGAACC	WT:0bp Targeted:341bp
	T004833-R1	ATATCCCCTTGTCCCTTCTGC	
②WT	T004833-F2	CAGCAAAACCTGGCTGTGGATC	WT:412bp Targeted:0bp
	T004833-R2	ATGAGGCCACCATGTGGGTGTC	

3. Gel Image & Conclusion





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 °C	5min	
2	98 °C	30s	20×
3	65 °C (-0.5 °C/cycle)	30s	
4	72 °C	45s	
5	98 °C	30s	20×
6	55 °C	30s	
7	72 °C	45s	
8	72 °C	5min	
9	10 °C	hold	