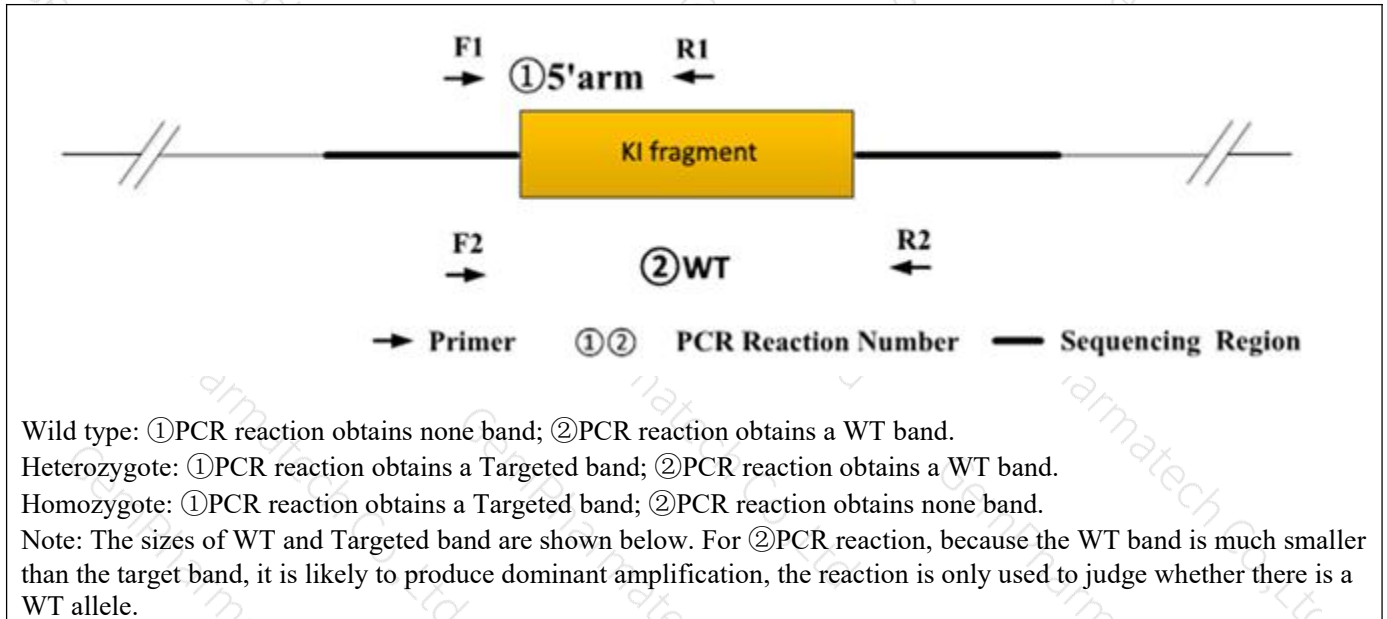


Genotyping Report

Strain ID	T005680	Strain Type	KI(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Dongdong Zhang	Gene Name	<i>H11-Nphs1-iCre</i>		

1. Strategy of Genotyping

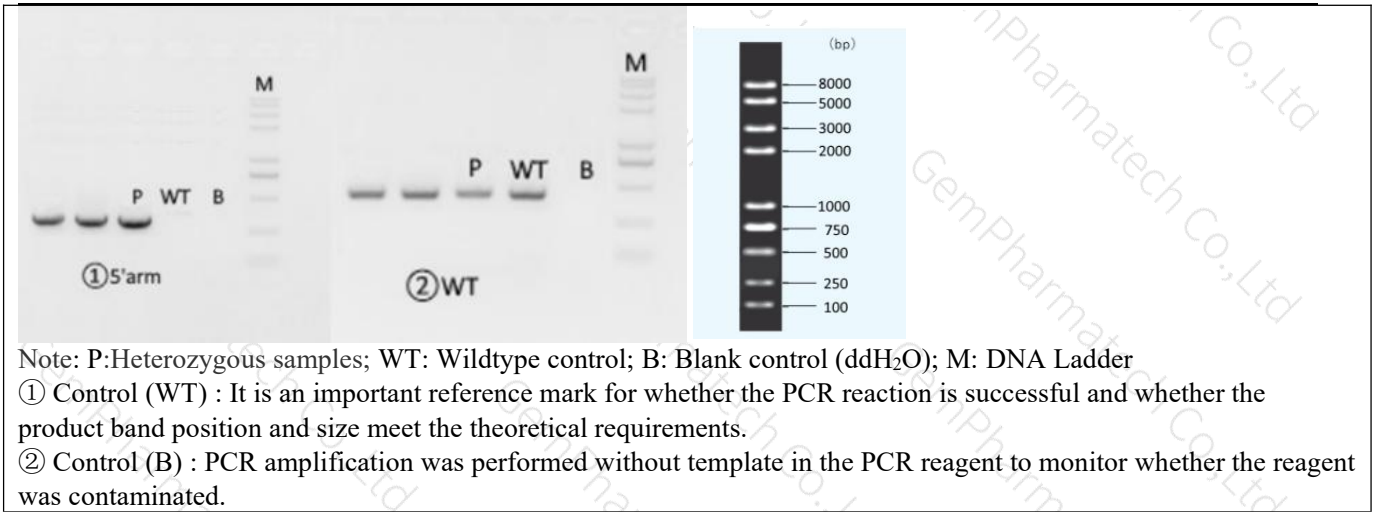


2. Primer Information

PCR No.	Primer No.	Primer Name	Sequence	Band Size
①5'arm	F1	H11-tF3	GGGCAGTCTGGTACTTCCAAGCT	WT:0bp Targeted:283bp
	R1	003778-Nphs1-5tR2	CCAAATTCATGGGTCTCAGAC	
②WT	F2	H11-wt-tF1	CAGCAAAACCTGGCTGTGGATC	WT:412bp Targeted:0bp
	R2	H11-wt-tR1	ATGAGCCACCATGTGGGTGTC	

3. Gel Image & Conclusion

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4. PCR Condition

PCR Reaction Component			
Seg.	reaction component	Volume (μl)	
1	2 × Rapid Taq Master Mix (Vazyme P222)	12.5	
2	ddH ₂ O	9.5	
3	Primer A(10pmol/μl)	1	
4	Primer B(10pmol/μl)	1	
5	Template(20~80ng/μl)	1	
PCR program I priority selection			
Seg.	Temp.	Time	Cycle
1	95℃	5min	
2	98℃	30s	20×
3	65℃* (-0.5℃/cycle)	30s	
4	72℃	45s*	
5	98℃	30s	15×
6	55℃*	30s	
7	72℃	45s*	
8	72℃	5min	
9	10℃	hold	
PCR program II the second choice			
Seg.	Temp.	Time	Cycle
1	95℃	5min	
2	98℃	30s	35×
3	58℃*	30s	

4	72℃	45s*	
5	72℃	5min	
6	10℃	hold	

Note*: Annealing temperature and extension time can be determined according to the actual amplification situation and amplification enzyme efficiency.