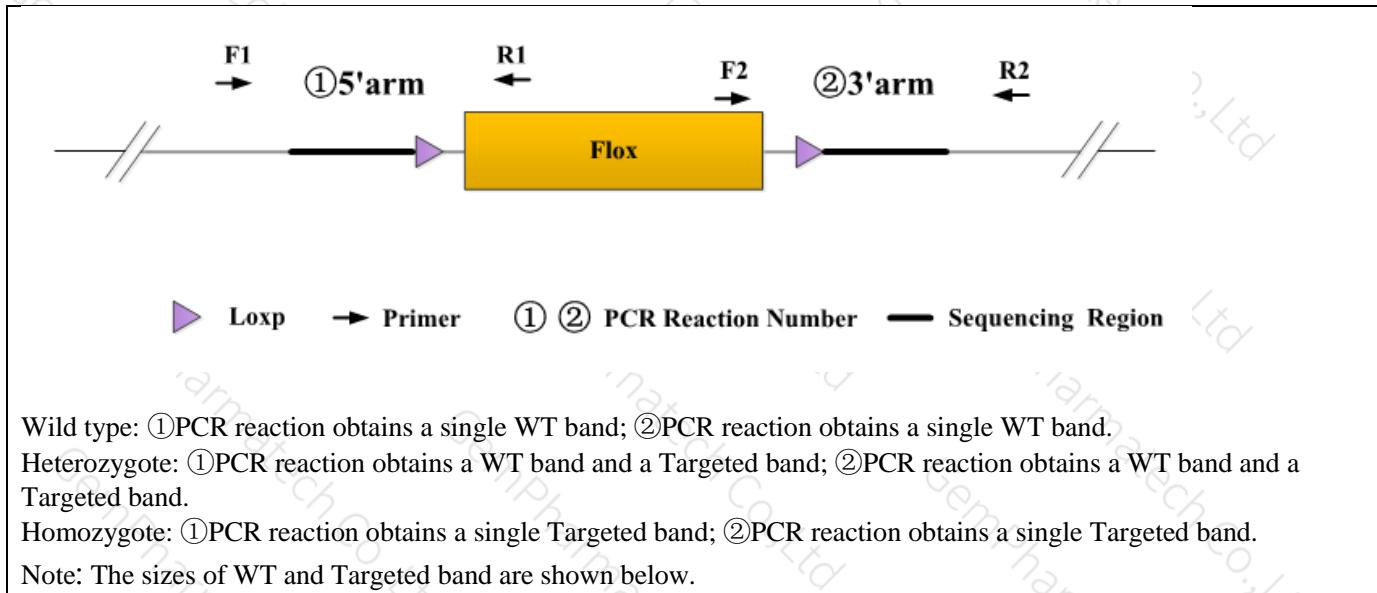




## Genotyping Report

Strain ID	T054498	Strain Type	CKO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Ya'nan Xu	Gene Name			<i>Foxk2</i>

### 1. Strategy of Genotyping



### 2. Primer Information

PCR No.	Primer No.	Sequence	Band Size
①(5'arm)	T054498-F1	ACACGACATTACACGCACATGCT	WT: 300bp Targeted:405bp
	T054498-R1	GCCAAGCTTCTGCATATGAGGAC	
②(3'arm)	T054498-F2	AGTGACTTCCAGTTTGTAAACTCAAGA	WT: 259bp Targeted:365bp
	T054498-R2	GTGAGGCTCAATGGACAGTACCA	

### 3. Gel Image & Conclusion





Note: P: Positive control; WT: Wildtype control; B: Blank control (ddH<sub>2</sub>O); M: DNA Ladder

① Control (WT) : It is an important reference mark for whether the PCR reaction is successful and whether the product band position and size meet the theoretical requirements.

② Control (B) : PCR amplification was performed without template in the PCR reagent to monitor whether the reagent was contaminated.

#### 4. PCR Condition

##### PCR Reaction Component

Seg.	reaction component	Volume (μl)
1	2 × Rapid Taq Master Mix (Vazyme P222)	12.5
2	ddH <sub>2</sub> O	9.5
3	Primer A(10pmol/μl)	1
4	Primer B(10pmol/μl)	1
5	Template(≈100ng/μl)	1

##### PCR program ① priority selection

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	

##### PCR program ② the second choice

Seg.	Temp.	Time	Cycle
1	95°C	5min	
2	98°C	30s	35×
3	58°C*	30s	
4	72°C	45s*	
5	72°C	5min	
6	10°C	hold	

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



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