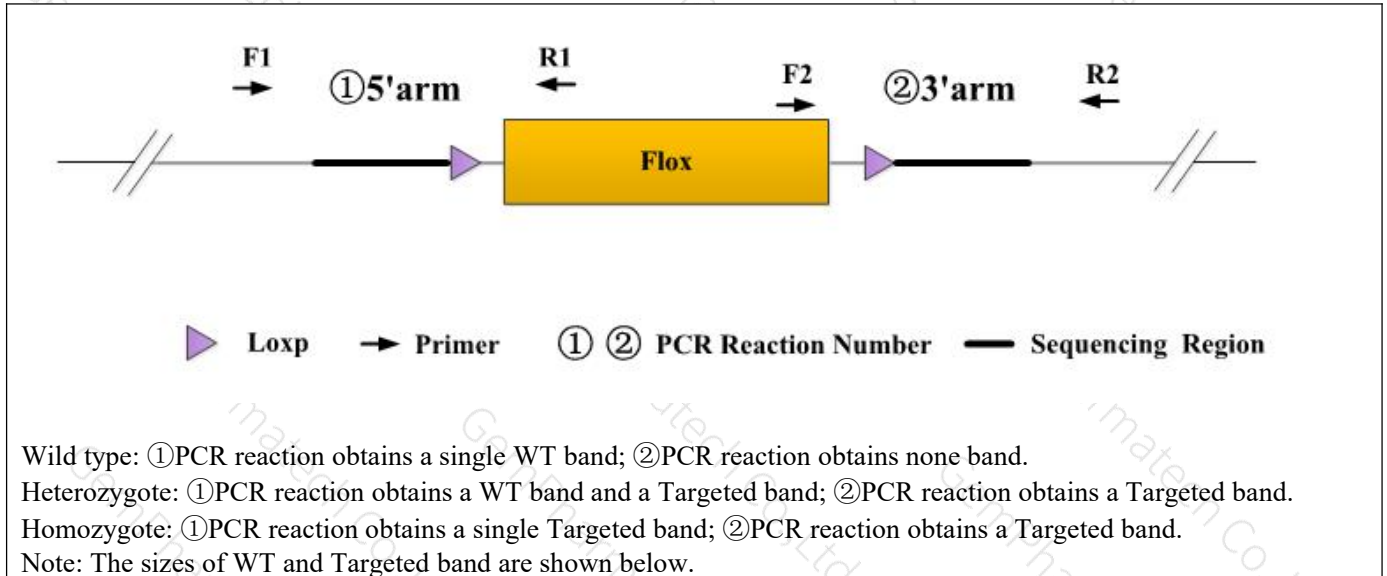


## Genotyping Report

Strain ID	T015943	Strain Type	CKO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Ya'nan Xu	Gene Name	<i>Scd1</i>		

### 1. Strategy of Genotyping



### 2. Primer Information

PCR No.	Primer No.	Primer Name	Sequence	Band Size
①(5'arm)	F1	T015943-F1B	TCACACAGTGGCAAGTGCTGA	WT:298bp Targeted:403bp
	R1	T015943-R1B	GGAATAACTCCCTCTGAACCAGGA	
②(3'arm)	F2	T015943-F2B	TCACCGTAAGTTCCTCAGGCAA	WT:351bp Targeted:457bp
	R2	T015943-R2B	TGGATGCGACAAGACATCCAGAG	

### 3. Gel Image & Conclusion



- 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

(Generally recommend to use Vazyme P222; If the sequences contain special structures such as GC% ≥ 60% or GC% ≤ 40%, recommend to use Vazyme P515.)

PCR Reaction Component		
Seg.	reaction component	Volume (μl)
1	2 × Rapid Taq Master Mix(Vazyme P222) or 2 × Phanta Max Master Mix (Vazyme P515)	12.5
2	ddH <sub>2</sub> 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°C	5min	
2	98°C	30s	20×
3	65°C* (-0.5°C/cycle)	30s	
4	72°C	45s*	
5	98°C	30s	
6	55°C*	30s	15×
7	72°C	45s*	
8	72°C	5min	
9	10°C	hold	

#### PCR program II 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

