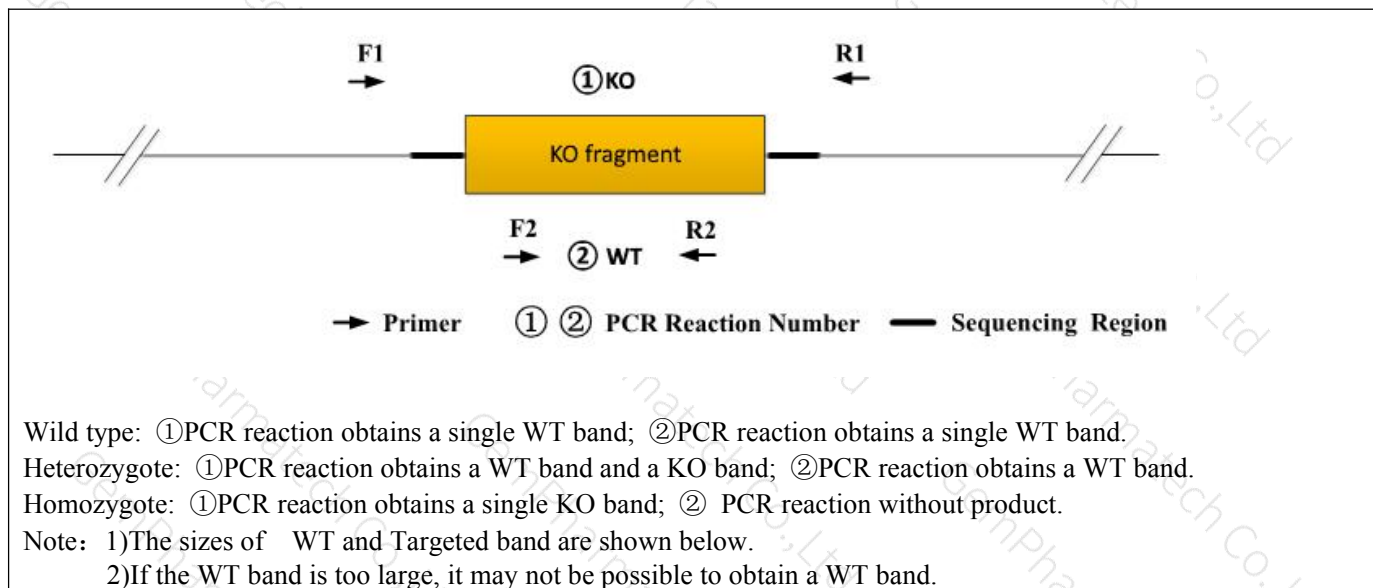


Genotyping Report

Strain ID	T027612	Strain Type	KO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Zifan Lin	Gene Name	Trpm8		

1. Strategy of Genotyping



2. Primer Information

PCR No.	Primer No.	Primer Name	Sequence	Band Size
PCR①	F1	JS08547-Trpm8-5wt-tF1	GTCTCTTGATGTTTTCCCATTTGGG	WT: 7094 bp KO: 223bp
	R1	JS08547-Trpm8-3wt-tR1	CTAGACACATGCACGTATAGTGATCTTG	
PCR②	F2	JS18547-Trpm8-wt-F1	CCTGTGAGACACTGTGTAGCCCA	WT: 330bp KO: 0 bp
	R2	JS18547-Trpm8-wt-R1	GAAGCACTCAGAGCTACGTCCACC	

3. Gel Image

ctacttatgatacatagctattgcagattccctca-----6875bp-----TCTTggccttgacttcatactcagcatcttctctct



Note: P: Positive control; W: Wildtype control; B: Blank control (ddH₂O); 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	20×
2	98℃	30s	
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	