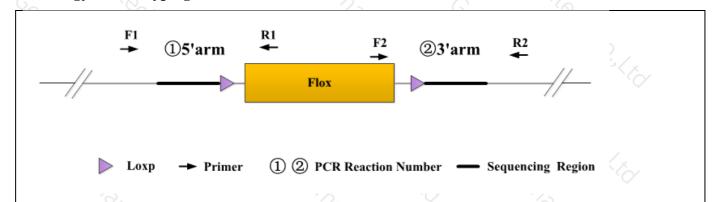


Genotyping Report

Strain ID	T052143	Strain Type	CKO(Cas9)	Genetic Background	C57BL/6JGpt
Designer	Ya'nan Xu	Gene Name	-3 </td <td>P2ry10</td> <th>0)</th>	P2ry10	0)

1. Strategy of Genotyping



Wild type: ①PCR reaction obtains a single WT band; ②PCR reaction obtains a single WT band.

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

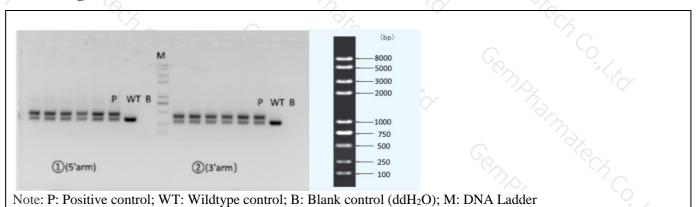
Homozygote: ①PCR reaction obtains a single Targeted band; ②PCR reaction obtains a single Targeted band.

Note: The sizes of WT and Targeted band are shown below.

2. Primer Information

1 100			7/1
PCR No.	Primer No.	Sequence	Band Size
①(5'arm)	T052143-F1	2143-F1 CACTCCTAACATTTCCAGAAGACCAC	
	T052143-R1	ACCTTCAACAAGGTCACATCTACCTC	Targeted:413bp
②(3'arm)	T052143-F2	TGGAACAGAAGATGGTTACAGTTCGT	WT: 285bp Targeted:391bp
	T052143-R2	ACCAGAAGCAGTCAGTGTTATAGGACC	

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

PCR Reaction Co	mponent) D	7) (V	
Seg.	reaction	reaction component		
1700	2 × Rapid Taq Master Mix (Vazy	2 × Rapid Taq Master Mix (Vazyme P222)		
2	ddH2O	70	9.5	
3	Primer A(10pmol/μl)	52 3/x	1	
4	Primer B(10pmol/μl)	Primer B(10pmol/μl)		
5	Template(≈100ng/μl)	Template(≈100ng/μl)		
PCR program ①	priority selection	C C	Sch 3/x	
Seg.	Temp.	Time	Cycle	
1	95℃	5min	19/77	
2	98℃	30s	20×	
3	65°C* (-0.5°C/cycle)	30s	30. 190	
4 %	72℃	45s*	3/	
5	98℃	30s	20×	
6	55℃*	30s	7,00	
7	72℃	45s*	%, ~~~~	
8	72℃	5min	3/x 3/x	
9	10°C	hold	J ³ , 4	
PCR program ②	the second choice	73x 60x	- CX	
Seg.	Temp.	Time	Cycle	
1 3/7	95℃	5min	1977 3/24	
2	98℃	30s	35×	
3	58℃*	30s	3	
4	72℃	45s*	S	
5	72℃	5min		
6	10℃	hold	79 ₀	

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