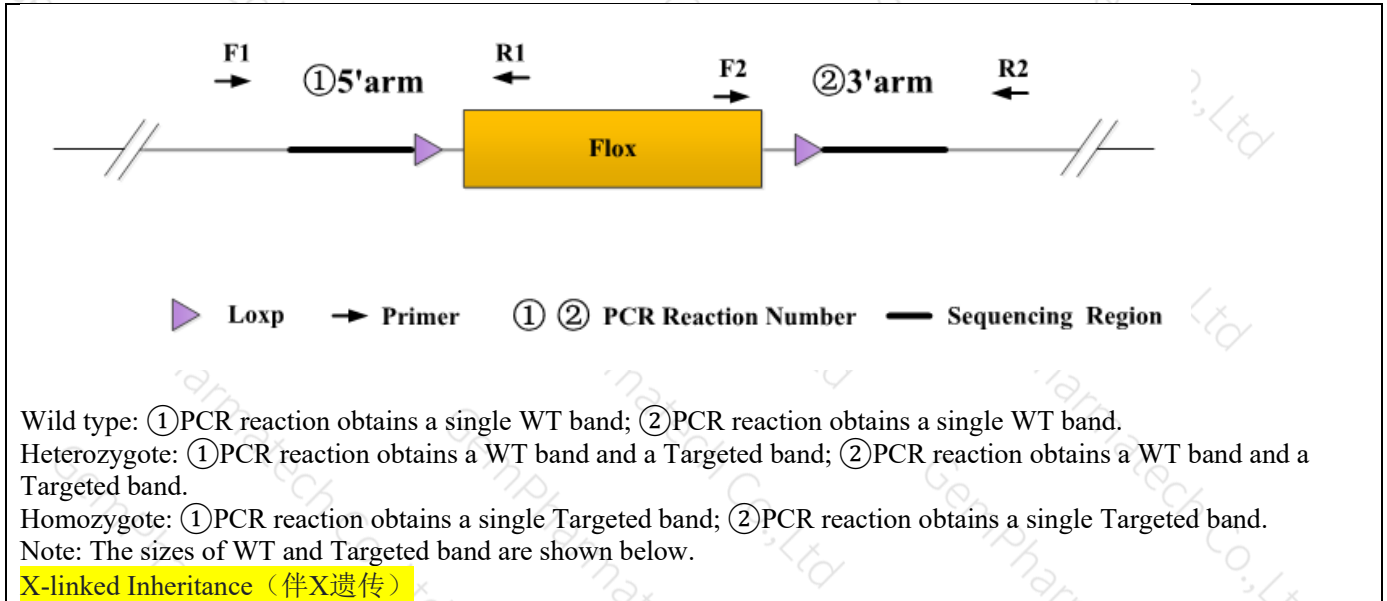


Genotyping Report

| | | | | | |
|-----------|-----------|-------------|-----------|--------------------|-------------|
| Strain ID | T052012 | Strain Type | CKO(Cas9) | Genetic Background | C57BL/6JGpt |
| Designer | Ya'nan Xu | Gene Name | Cxcr3 | | |

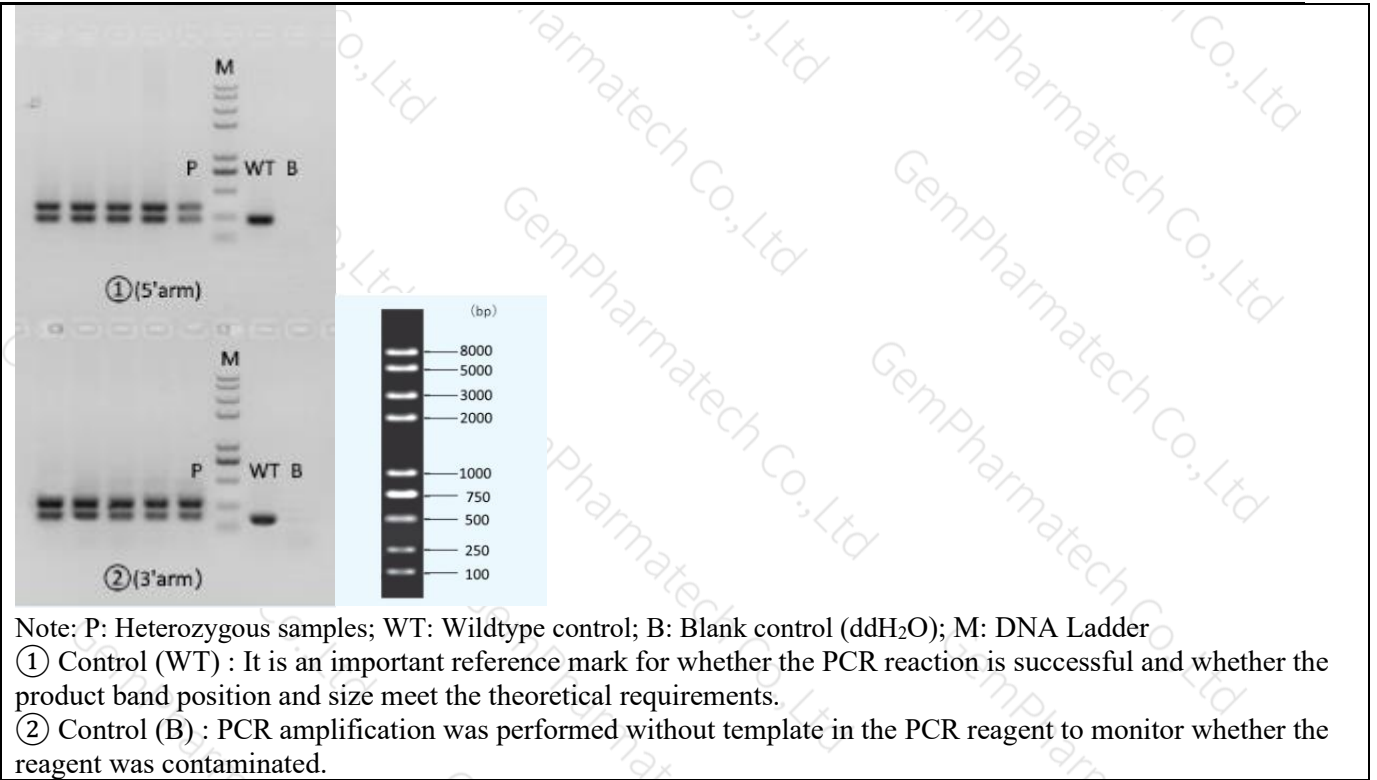
1. Strategy of Genotyping



2. Primer Information

| PCR No. | Primer No. | Sequence | Band Size |
|----------|------------|--------------------------|-----------------------------|
| ①(5'arm) | T052012-F1 | AGGAAGTCAGGGTCACCAGATAAG | WT:227bp Targeted:333bp |
| | T052012-R1 | TATCATCCAGGACAGGGATTCTC | |
| ②(3'arm) | T052012-F2 | CTTCCAGAAGGGTAGGTGACAG | WT:167 bp Targeted:268bp |
| | T052012-R2 | GCAAACCTTACCCAGCTAACTGCT | |

3. Gel Image & Conclusion



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 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°C | 5min | 20× |
| 2 | 98°C | 30s | |
| 3 | 65°C*(-0.5°C/cycle) | 30s | |
| 4 | 72°C | 45s* | |
| 5 | 98°C | 30s | 15× |
| 6 | 55°C* | 30s | |

| 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 and amplification enzyme efficiency.

Reviewer:Ting Sun

Date:2021-7-18