

PCR protocol

RBRC 02736

Strain name C57BL/6J-Tg(Dsg3TCR162)3

| Primer | primer_name | primer_seq | length | |
|--------|-------------|--------------------------------------|--------|-------------------------|
| 1 | | gcccgggGGAGAGATAACTCAAAGCTTCAGAGAAG | 36 | Depositor's primer |
| 2 | | gcccgggCCCACCCAGTGCATGCATACCTCAGAGA | 35 | Depositor's primer |
| 3 | | gcccgggTCCAAGCTGTGGGCCACACCAGTGAATTT | 36 | Depositor's primer |
| 4 | | gctcgagTAGTTCTGAGATGGGCTCCAGACTCTT | 34 | Depositor's primer |
| 5 | oIMR0042 | CTAGGCCACAGAATTGAAAGATCT | 24 | JAX Genotyping Protocol |
| 6 | oIMR0043 | GTAGGTGGAAATTCTAGCATCATCC | 25 | JAX Genotyping Protocol |
| | | | | |
| | | | | |
| | | | | |

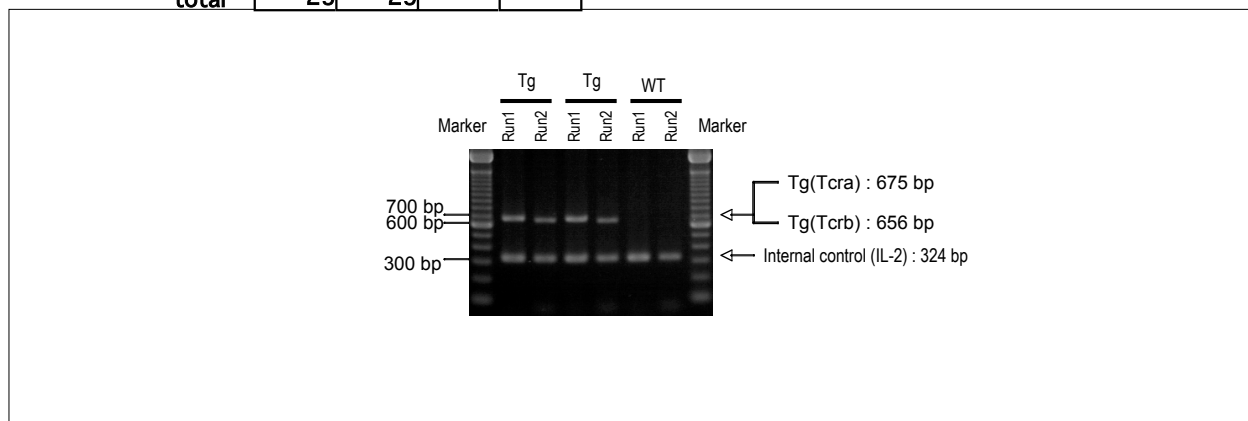
Reaction Components

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

Reaction Conditions

| step | temp (°C) | time (sec) | note |
|------|-----------|------------|-----------------------|
| 1 | 94 | 180 | |
| 2 | 94 | 30* | |
| 3 | 60 | 30* | |
| 4 | 72 | 60* | *Repeat for 30 cycles |
| 5 | 72 | 300 | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |

| | Conc | Run 1 | Run 2 | Run 3 | Run 4 |
|------------|------------------|-------|-------|-------|-------|
| H2O | | 15.3 | 15.3 | | |
| PCR buffer | 10 X | 2.5 | 2.5 | | |
| dNTP | 2.5 mM | 2.0 | 2.0 | | |
| primer 1 | 10 pmol/ μ l | 1.0 | 0 | | |
| primer 2 | 10 pmol/ μ l | 0 | 1.0 | | |
| primer 3 | 10 pmol/ μ l | 1.0 | 0 | | |
| primer 4 | 10 pmol/ μ l | 0 | 1.0 | | |
| primer 5 | 10 pmol/ μ l | 1.0 | 1.0 | | |
| primer 6 | 10 pmol/ μ l | 1.0 | 1.0 | | |
| DNA | | 1.0 | 1.0 | | |
| Taq | | 0.2 | 0.2 | | |
| | | | | | |
| | | | | | |
| | | | | | |
| total | | 25 | 25 | | |



Marker
100 bp ladder (Invitrogen, Cat. 15628-050)

Gel
2% agarose in 1 X TAE (100 V, 30 min)

| PCR products | | product_size | product |
|--------------|------------|--------------|-------------------------|
| primer_set | primer_set | | |
| primer 1 | primer 3 | 675 bp | Tg (alpha-chain) |
| primer 2 | primer 4 | 656 bp | Tg (beta-chain) |
| primer 5 | primer 6 | 324 bp | Internal control (IL-2) |
| | | | |
| | | | |

Comment