# PCR protocol

**RBRC** 02655 **Strain name** B6.Cg-Xist<tm5Sado>

**Primer**

| | primer_name | primer_seq | length | | |
|---|---|---|---|---|---|
| 1 | R700P2 | CGGGGCTTGGTGGATGGAAAT | 21 | | Depositor's primer |
| 2 | F1063AS | GCACAACCCCGCAAATGCTA | 20 | | Depositor's primer |
| 3 | dA1F | GCAGGTCGAGGGACCTAATA | 20 | | Depositor's primer |

**Reaction Components**

**Enzyme** TaKaRa Ex Taq (Cat. No. RR001A)

| | Conc | Run 1 | Run 2 | Run 3 | Run 4 |
|---|---|---|---|---|---|
| H2O | | 16.3 | | | |
| PCR buffer | 10 X | 2.5 | | | |
| dNTP | 2.5 mM | 2.0 | | | |
| primer 1 | 10 pmol/μl | 1.0 | | | |
| primer 2 | 10 pmol/μl | 1.0 | | | |
| primer 3 | 10 pmol/μl | 1.0 | | | |
| DNA | | 1.0 | | | |
| Taq | | 0.2 | | | |
| | | | | | |
| | | | | | |
| | | | | | |
| | | | | | |
| | | | | | |
| | total | 25 | | | |

**Reaction Conditions**

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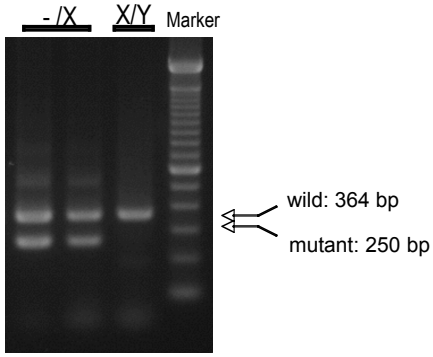


**Marker**

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

**Gel**

2% agarose in 1 X TAE (100 V, 35 min)

**PCR products**

| primer_set | | product_size | product |
|---|---|---|---|
| primer 1 | primer 2 | 364 bp | WT |
| primer 3 | primer 2 | 250 bp | KO |
| | | | |
| | | | |
| | | | |

**Comment**