



RBRC 00462      Strain name MAK Knockout mice B6

| Primer | primer_name | primer_seq              | length |                    |
|--------|-------------|-------------------------|--------|--------------------|
| 1      | SCR4        | AGCCATCTCGCCAGCACCTCATA | 23     | Depositor's primer |
| 2      | G3          | GGGCCAGCTCATTCTCCACTC   | 22     | Depositor's primer |
| 3      | MAK-R       | AGCACGAAAGTGCCAGCAACTC  | 24     | Depositor's primer |
|        |             |                         |        |                    |
|        |             |                         |        |                    |
|        |             |                         |        |                    |
|        |             |                         |        |                    |
|        |             |                         |        |                    |
|        |             |                         |        |                    |
|        |             |                         |        |                    |

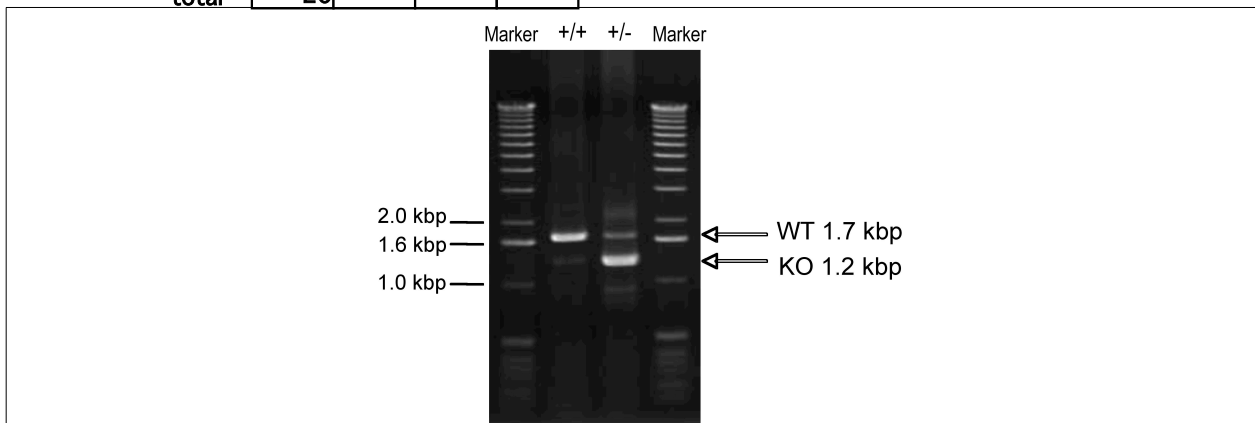
**Reaction Components**

Enzyme TaKaRa LA Taq with GC buffer (Cat. No. RR02AG)

**Reaction Conditions**

|              | Conc             | Run 1     | Run 2 | Run 3 | Run 4 |
|--------------|------------------|-----------|-------|-------|-------|
| H2O          |                  | 3.9       |       |       |       |
| PCR buffer   | 2 X GC I         | 10.0      |       |       |       |
| dNTP         | 2.5 mM           | 2.0       |       |       |       |
| primer 1     | 10 pmol/ $\mu$ l | 1.0       |       |       |       |
| primer 2     | 10 pmol/ $\mu$ l | 1.0       |       |       |       |
| primer 3     | 10 pmol/ $\mu$ l | 1.0       |       |       |       |
| DNA          |                  | 1.0       |       |       |       |
| Taq          |                  | 0.1       |       |       |       |
|              |                  |           |       |       |       |
|              |                  |           |       |       |       |
|              |                  |           |       |       |       |
|              |                  |           |       |       |       |
|              |                  |           |       |       |       |
| <b>total</b> |                  | <b>20</b> |       |       |       |

| step | temp (°C) | time (sec) | note                  |
|------|-----------|------------|-----------------------|
| 1    | 95        | 180        |                       |
| 2    | 95        | 30*        |                       |
| 3    | 66        | 30*        |                       |
| 4    | 72        | 90*        | *Repeat for 30 cycles |
| 5    | 72        | 300        |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |
|      |           |            |                       |



**Marker**  
1 kb DNA ladder (Invitrogen, Cat. 15615-024)

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

| PCR products |          | product_size | product |
|--------------|----------|--------------|---------|
| primer_1     | primer_2 |              |         |
| primer 1     | primer 2 | 1.2 kbp      | KO      |
| primer 1     | primer 3 | 1.7 kbp      | WT      |
|              |          |              |         |
|              |          |              |         |
|              |          |              |         |
|              |          |              |         |

Comment