# PCR protocol

Sheet ID PS_06580
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**RBRC** 09428 **Strain name** C57BL/6-Adarb1<flox/flox>/VAChT-Cre. Fast mouse

| Primer | primer_name | primer_seq | length | | |
|---|---|---|---|---|---|
| 1 | OYC52-9 | GCTGTGCATAAAGAAACCTGAGCAG | 25 | | Depositor's primer |
| 2 | OYC52-8 | CTGGTTCATAACAGATCCTCAGGG | 24 | | J Neurosci., 30, 11917-11925 (2010). |
| 3 | OYC52-7 | GTCTCCCTTGTCCTTCCAGGTAGC | 24 | | J Neurosci., 30, 11917-11925 (2010). |
| 4 | promoter | GCGGAGATCGCAAGCCTCTGG | 21 | | RIKEN BRC |
| 5 | | CGAGTTGATAGCTGGCTGG | 19 | | Depositor's primer |

## Reaction Components

**Enzyme**: TOYOBO KOD FX (Code.No : KFX-101)

| | Conc | Run 1 | Run 2 | Run 3 | Run 4 |
|---|---|---|---|---|---|
| H2O | | 2.3 | | | |
| PCR buffer | 2 X | 10.0 | | | |
| dNTP | 2.0 mM | 4.0 | | | |
| primer 1 | 10 pmol/μl | 0.5 | | | |
| primer 2 | 10 pmol/μl | 0.5 | | | |
| primer 3 | 10 pmol/μl | 0.5 | | | |
| primer 4 | 10 pmol/μl | 0.5 | | | |
| primer 5 | 10 pmol/μl | 0.5 | | | |
| DNA | | 1.0 | | | |
| Taq | 1 U/μl | 0.2 | | | |
| | | | | | |
| | | | | | |
| | | | | | |
| | | | | | |
| total | | 20 | | | |

## Reaction Conditions

| step | temp (℃) | time (sec) | | note |
|---|---|---|---|---|
| 1 | 94 | 120 | | |
| 2 | 98 | 10 | * | |
| 3 | 60 | 30 | * | |
| 4 | 68 | 180 | * | *Repeat for 30 cycles |
| | | | | |
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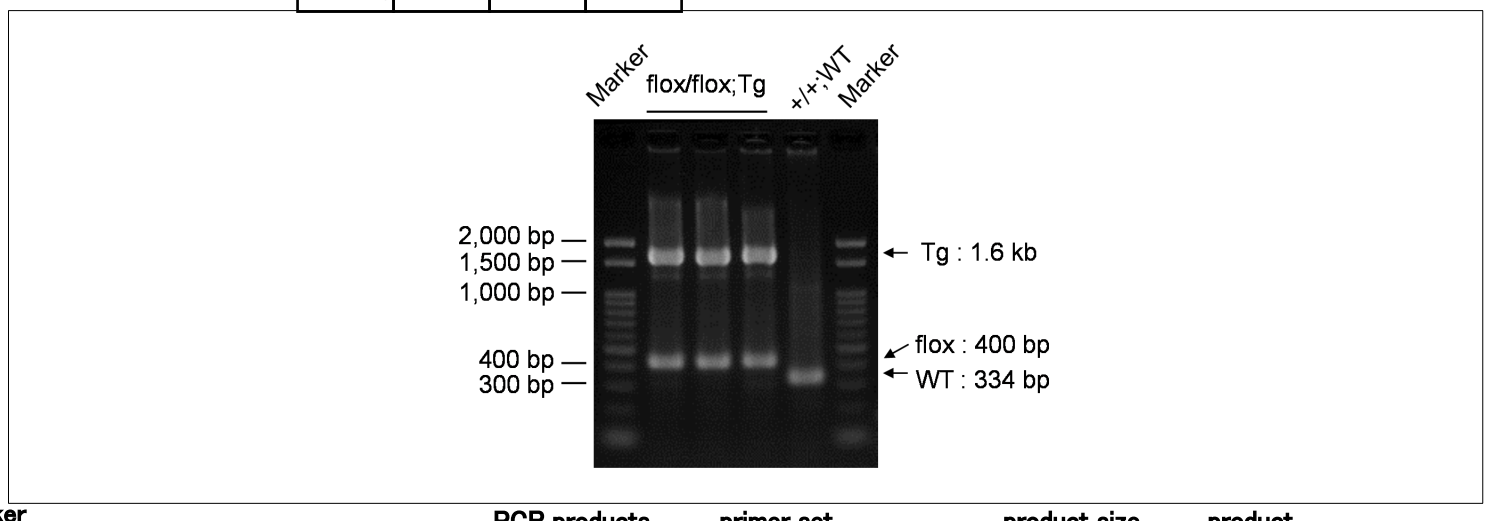


**Marker**: Gene Ladder 100 (Wako, Cat. 316-06951)

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

| PCR products | primer_set | product_size | product |
|---|---|---|---|
| primer 1 | primer 2 | 334 bp | WT |
| primer 1 | primer 2 | 400 bp | flox |
| primer 4 | primer 5 | 1.6 kb | Tg |
| primer 1 | primer 3 | | null |
| | | | |
| | | | |

**Comment**

Novagen KOD Xtreme™ Hot Start DNA Polymerase can be purchased from EMD.