

B6.Cg-Il17rc^{tm1.1Koll}/J

Stock No: 031002

Protocol 32047: Standard PCR Assay - Il17rc^{tm1.1Koll}

Version 1.0

Notes

The genotyping protocol(s) presented here have been optimized for reagents and conditions used by The Jackson Laboratory (JAX). To genotype animals, JAX recommends researchers validate the assay independently upon receipt of animals into their facility. Reaction cycling temperature and times may require additional optimization based on the specific genotyping reagents used.

Expected Results

>[chr6:113472437+113472662](#) 226bp AGGCATGAGGATTGCAGACT CAGACTTTCAGCTTCTTCAGG

Mutant = ~275 bp

Heterozygote = ~275 bp and 226 bp

Wild type = 226 bp

JAX Protocol

Protocol Primers

| PRIMER | 5' LABEL | SEQUENCE 5' → 3' | 3' LABEL | PRIMER TYPE | REACTION | NOTE |
|--------|----------|-------------------------------|----------|-------------|----------|------|
| 38239 | | AGG CAT GAG GAT TGC AGA CT | | Forward | A | |
| 38240 | | CAG ACT TTC CAG CTT CTT CAG G | | Reverse | A | |

Reaction A

| COMPONENT | FINAL CONCENTRATION |
|----------------------|---------------------|
| ddH ₂ O | |
| Kapa 2G HS buffer | 1.30 X |
| MgCl ₂ | 2.60 mM |
| dNTPS-kapa | 0.26 mM |
| 38239 | 0.50 uM |
| 38240 | 0.50 uM |
| Glycerol | 6.50 % |
| Dye | 1.00 X |
| Kapa 2G HS taq polym | 0.03 U/ul |
| DNA | |

Cycling

| STEP | TEMP °C | TIME | NOTE |
|------|---------|------|--|
| 1 | 94.0 | -- | |
| 2 | 94.0 | -- | |
| 3 | 65.0 | -- | -0.5 C per cycle decrease |
| 4 | 68.0 | -- | |
| 5 | | -- | repeat steps 2-4 for 10 cycles (Touchdown) |
| 6 | 94.0 | -- | |
| 7 | 60.0 | -- | |
| 8 | 72.0 | -- | |
| 9 | | -- | repeat steps 6-8 for 28 cycles |
| 10 | 72.0 | -- | |
| 11 | 10.0 | -- | hold |

JAX uses a very high speed Taq (~1000 bp/sec), use cycling times recommended for your reagents.

JAX uses a 'touchdown' cycling protocol and therefore has not calculated the optimal annealing temperature for each set of primers.

