

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	36902
MMRRC Strain Name	B6.Cg-Tg(IL2-emGFP,IL21-mCherry)6Wjl/Mmnc
Gene Name(s)	interleukin 2 (IL2) interleukin 21 (IL21)
Breeding Protocol(s)	Backcross to C57BL/6J
Protocol Date	9/23/13

MMRRC #36902 PCR Reaction

Thermal Cycler:

Step 1: 94°C for 5 min
 Step 2: 94°C for 30 sec
 Step 3: 57°C for 30 sec
 Step 4: 72°C for 30 sec
 Step 5: Step 2 to 4; Cycles: 35
 Step 6: 72°C for 7 min

Taq: Apex and Chromataq 5X Buffer

	<u>1X</u>
ddH ₂ O	13
5X Buffer	5
25mM MgCl ₂	2
10mM dNTPs	0.5
10 μM Forward Primer	1
10 μM Reverse Primer	1
Taq	0.5
DNA	2

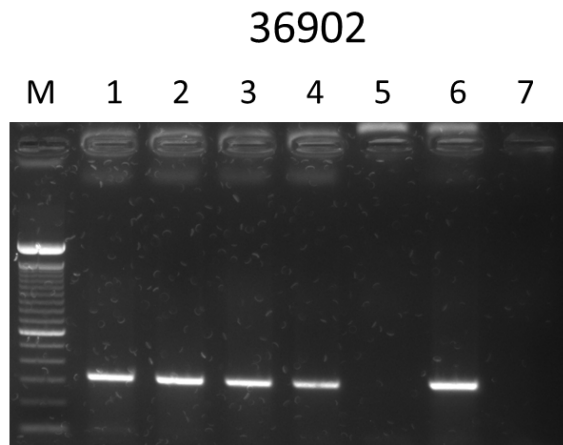
Primer sequences 5' to 3': Primers are 10 μM with respect to each primer.

pmcherry267F: CCTGTCCCCTCAGTTCATGT
 pmcherry591R: CTTGAGCTTCAGCCTCTGCT

Bands expected:

MUTANT: 320 bp
 WT: No Band

Run on 2.0% agarose gel in TAE.



Lane 1 to 4, 6: Mut; Lane 5: WT; Lane 7: H₂O;
 M: 100 bp DNA ladder (Invitrogen)