

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	15749
MMRRC Strain Name	B6;CBA-Tg(Axin2-d2EGFP)5Cos/Mmnc
Gene Name(s)	Axin2; Tg(Axin-d2EGFP)5Cos
Breeding Protocol(s)	Backcross to C57BL/6J
Protocol Date	9/30/13

MMRRC# 15749 PCR Reaction

	<u>1X</u>
DNA	1.0
5X Buffer	5.0
10mM dNTPs	1.0
20 uM Primer F/R	1.0
Taq	1.0
ddH ₂ O	14.5
25mM MgCl ₂	1.5

Thermal Cycler:

94°C for 45 sec
 58°C for 45 sec
 72°C for 120 sec
 35x

Taq: **Apex and Chromataq 10X Buffer**

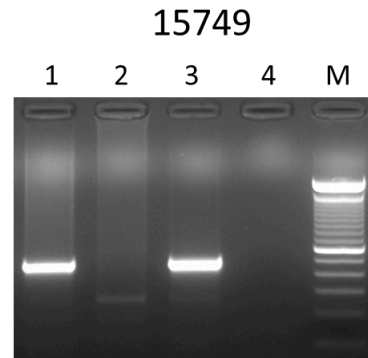
Bands: MUTANT: 478bp
 WT: no band

Primer sequences 5' to 3': Primers are 10uM with respect to each primer (20uM total).

TL-2A : 5' GCC TTT GAA AAA GCT GCG TCG 3' 21bp

d2EGFP1 : 5' TGT GGT CGG GGT AGC GGC TG 3' 20bp

Run on 2.0% agarose gel in TAE.



Lane 1, 3: Mut; Lane 2: WT; Lane 4: H₂O
 M: 100 bp DNA ladder (Invitrogen)