

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	229
MMRRC Strain Name	129S-In(11Trp53;11Wnt3)8Brd/Mmnc
Gene Name(s)	Inversion-transformation related protein 53 (Trp53), Wingless-related MMTV integration site 3 (Wnt3)
Breeding Protocol(s)	Sib-mating
Protocol Date	10/7/13

MMRRC #229 PCR Reaction

	<u>1X</u>
ddH ₂ O	13
5X Buffer	5.0
25mM MgCl ₂	2
10mM dNTPs	0.5
10 μM Primer F	1
10 μM Primer R	1
Taq	0.5
DNA	2

Thermal Cycler:

Step 1: 94°C for 5 min
 Step 2: 94°C for 30 sec
 Step 3: 51°C for 30 sec
 Step 4: 72°C for 45 sec
 Step 5: 35x from step 2 to step 4
 Step 6: 72°C for 7 min

Taq: Apex and Chromataq 5X Buffer

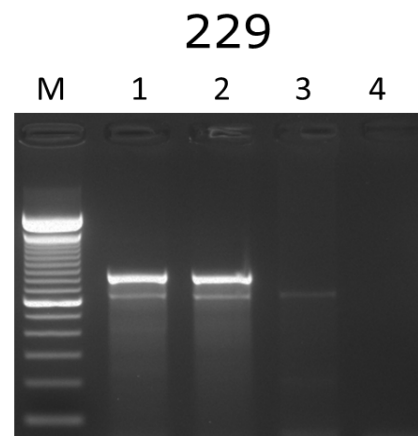
Primer sequences 5' to 3': Primers are 10 μM with respect to each primer.

P3-HPRT (229): AGGATGTGTTACGTGGAAGA

HPRT-R (229): CCAGTTTCACTAATGACACA

Bands expected: Tg+ ~800bp

Run on 2% agarose gel in TAE.



Lane 1, 2: Tg+; Lane 3: WT; Lane 4: H₂O
 M: 100 bp DNA ladder Invitrogen