

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	30969
MMRRC Strain Name	B6.129P2- <i>Gria3</i> ^{tm1Dgen} /Mmnc
Gene Name(s)	Gria3
Breeding Protocol(s)	Sib-mate or Backcross to C57BL/6J
Protocol Date	10-28-13

MMRRC# 30969 PCR Reaction

NOTE: GRIA3 is x-linked

Initial: 94°C for 7 min

Denaturing: 94°C for 45 sec

Annealing Temp.: 60°C for 45 sec

Extension: 72°C for 90 sec

Cycles: 35

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

	<u>1X</u>
DNA	1.0
10X Buffer	5.0
10mM dNTPs	0.5
20 uM Primer 1&2&3	1.5
Taq	0.5
ddH ₂ O	15.0
25mM MgCl ₂	2.0

Bands: MUTANT: 380bp

WT: 275bp

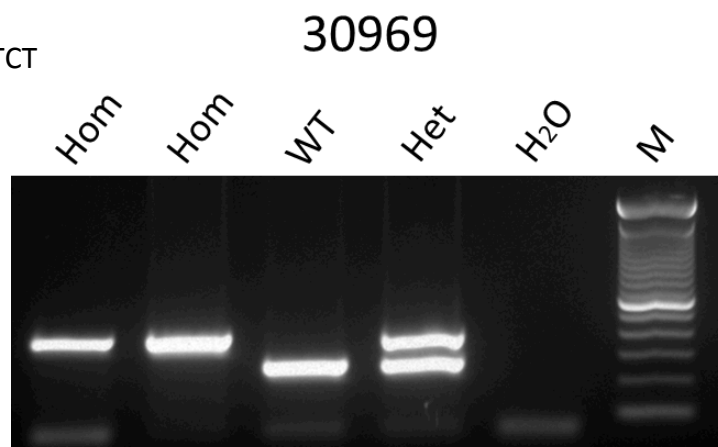
Primer sequences 5' to 3': Primers are 10uM with respect to each primer (20uM total).

GS1 - AGCTGATATAGCTGTTGCTCCACTC

GS2 - GGTCACGAGGTTCTTCATTGTTGTC

Neo - GGGTGGGATTAGATAAATGCCTGCTCT

Run on 2.0% agarose gel in TAE.



M: 100 bp DNA ladder (Invitrogen)