

## MMRRC UNC – Genotyping Protocol

<b>MMRRC Strain ID</b>	30658
<b>MMRRC Strain Name</b>	STOCK <i>Grin2c</i> <sup>tm1Bnno</sup> /Mmnc
<b>Gene Name(s)</b>	glutamate receptor, ionotropic, NMDA2C ( <i>Grin2c</i> )
<b>Breeding Protocol(s)</b>	Intra-Strain Mating
<b>Protocol Date</b>	10/16/13

### MMRRC #30658 PCR Reaction

	<u>1X</u>
ddH <sub>2</sub> O	13
5X Buffer	5
25mM MgCl <sub>2</sub>	2
10mM dNTPs	0.5
10uM WT Primer	1
10uM Mut Primer	1
Taq	0.5
DNA	2

**NOTE: Set up two, 2-primer reactions. Primer pairs are: (b-gal.2F & b-gal.4R) for the mutant reaction and (NR2C.1F & NR2C.2R) for the wild-type reaction**

#### **Thermal Cycler:**

Step 1: 94°C for 5 min

Step 2: 94°C for 30 sec

Step 3: 63°C for 45 sec

Step 4: 72°C for 60 sec

Step 2 to 4 Cycles: 33

Step 5: 72°C for 7 min

**Taq: Apex Taq and Chromataq 5X Buffer**

#### **Bands:**

MUTANT: ~300bp

WT: ~360bp

**Primer sequences 5' to 3':**

b-gal.2F: 5'- GGAAAGCTGGCTGGAGTGCATCTT -3'

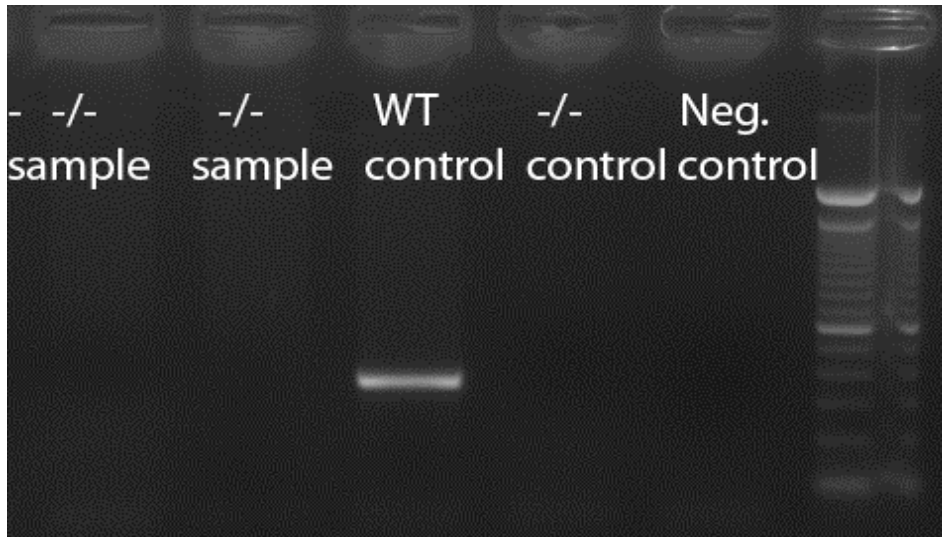
b-gal.4R: 5'- GCAAACGACTGTCCTGGCCGTAAC -3'

NR2C.1F: 5'-TCTCACTTCACTCCTTGGTGC-3'

NR2C.2R: 5'-GTGAGGACCACAGCAGAACCT C- 3'

Run on 2% agarose gel.

Representation of a wild-type reaction ran 2% agarose gel:



Representation of a mutant reaction ran on a 2% agarose gel:

