

## MMRRC UNC –Genotyping Protocol

<b>MMRRC Strain ID</b>	10602
<b>MMRRC Strain Name</b>	129S6/SvEvTac-Tg(Alb1-Ren)2Unc/Mmnc
<b>Gene Name(s)</b>	transgene insertion 2, University of North Carolina (Tg(Alb1-Ren)2Unc)
<b>Breeding Protocol(s)</b>	Intra-Strain Mating
<b>Protocol Date</b>	11/25/13

### MMRRC #10602 PCR Reaction

	<u>1X</u>
ddH <sub>2</sub> O	13
5X Buffer	5.0
25 mM MgCl <sub>2</sub>	2
10 mM 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: 60°C for 30 sec  
 Step 4: 72°C for 45 sec  
 Step 5: 35 x 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.

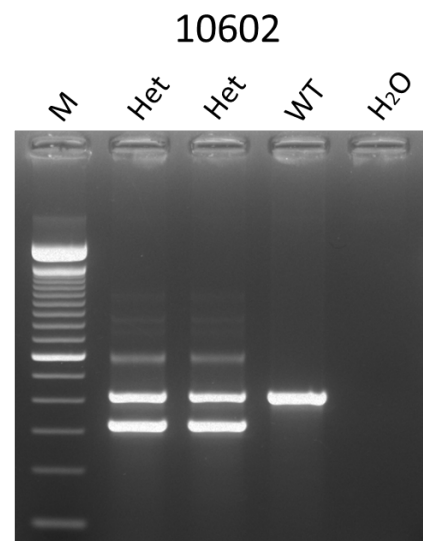
P1 (10602): TGGGATTCTAACCTGAGGACC

P2 (10602): CACAGATTGTAAGTCAAATCTGTCTG

P3 (10602): GT TCT TCTGAGGGGATCGGC

**Bands expected:** WT: 425 bp  
 MUTANT: 330 bp

Run on 2% agarose gel in TAE.



M: 100 bp DNA ladder (Invitrogen)