

## MMRRC UNC – Genotyping Protocol

<b>MMRRC Strain ID</b>	36801
<b>MMRRC Strain Name</b>	STOCK Tg(ACTB-HistH2b/GFP)1Mboi/Mmnc
<b>Gene Name(s)</b>	Histone H2b (HistH2b) actin, beta (ACTB) Green Fluorescent Protein (GFP)
<b>Breeding Protocol(s)</b>	Backcross to C57BL/6J
<b>Protocol Date</b>	9/23/13

### MMRRC #36801 PCR Reaction

**Thermal Cycler:**

Step 1: 94°C for 5 min  
 Step 2: 94°C for 60 sec  
 Step 3: 52°C for 60 sec  
 Step 4: 72°C for 90 sec  
 Step 5: Step 2 to 4; Cycles: 35  
 Step 6: 72°C for 7 min

	<b><u>1X</u></b>
ddH <sub>2</sub> O	13
5X Buffer	5
25 mM MgCl <sub>2</sub>	2
10 mM dNTPs	0.5
10 μM Forward Primer	1
10 μM Reverse Primer	1
Taq	0.5
DNA	2

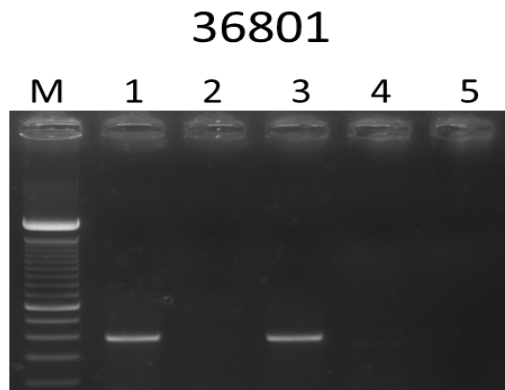
**Taq: Apex and Chromataq 5X Buffer**

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

Forward (36801): AGTCCTGAGGGATACGTGC  
 Reverse (36801): GACCATGTGATCTCTTTCC

**Bands expected:** MUTANT: 438 bp  
 WT: No Band

Run on 1.0% agarose gel in TAE.



Lane 1, 3: Mut; Lane 2, 4: WT; Lane 5: H<sub>2</sub>O;  
 M: 100 bp DNA ladder (Invitrogen)