

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	30494
MMRRC Strain Name	B6;129S4- <i>Tmem123</i> ^{tm1Shr} /Mmnc
Gene Name(s)	Transmembrane protein 123 (Tmem123)
Breeding Protocol(s)	Intra-strain Mating
Protocol Date	8/12/13

MMRRC #30494 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: 53°C for 30 sec
 Step 4: 72°C for 1 min
 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.

O-2033: TCCCTAGAAAGCTTCCTAGAATCG

O-2035: AATTCCATGGTCCATTTCTAGCA

O-2037: TTGACAACTGTAACGGGTACGTAA

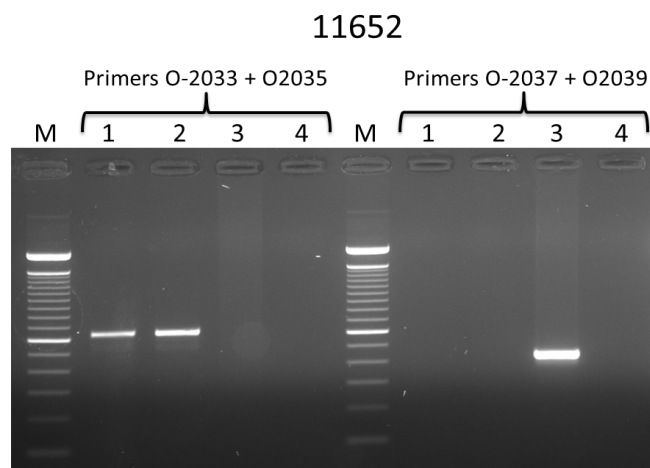
O-2039: GGTGAGCACTGCCTAACTATGGTA

Bands expected:

(Primers O-2033 + O2035) Mut 643 bp

(Primers O-2037 + O2039) WT 433 bp

Run on 1% agarose gel in TAE.



Lane 1, 2: Hom; Lane 3: WT; Lane 4: H₂O; M: 50 bp DNA ladder (Invitrogen)