

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	16994
MMRRC Strain Name	B6;129S-Myh10 ^{tm4Rsad} /Mmnc
Gene Name(s)	Myosin, heavy polypeptide 10, non-muscle (Myh10)
Breeding Protocol(s)	Sib-mating
Protocol Date	3/24/14

MMRRC #16994 PCR Reaction

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: 32x from step 2 to step 4
- Step 6: 72°C for 7 min

Taq: Apex and Chromataq 5X Buffer

	1X
ddH ₂ O	12
5 × Buffer	5.0
25 mM MgCl ₂	2
10 mM dNTPs	0.5
10 μM P1 (16994)	1
10 μM P2 (16994)	1
10 μM P3 (16994)	1
Taq	0.5
DNA	2

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

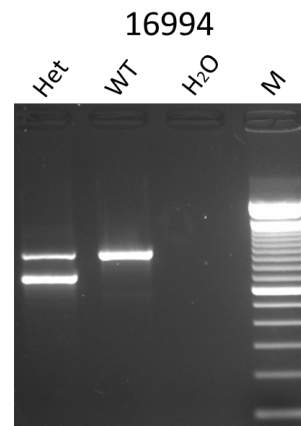
P1 (16994): GGCAAATGGGAATTATGGCT

P2 (16994): CTCTGGTCTGTGGTTTGTTC

P3 (16994): TTCGCAGCGCATCGCCTTCTA

Bands expected: WT: 931 bp
 MUTANT: 700 bp

Run on 2 % agarose gel in TAE.



M: 100 bp DNA ladder (Invitrogen)