

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	42051
MMRRC Strain Name	B6.129S1- <i>Atp6v1b1</i> ^{tm1Rpl} /Mmnc
Gene Name(s)	ATPase, H+ transporting, lysosomal V1 subunit B1
Breeding Protocol(s)	Random intra-strain mating
Protocol Date	9/22/2020

PCR Reaction

	<u>1X</u>
ddH ₂ O	11
5X Buffer	5
25mM MgCl ₂	2
10mM dNTPs	0.5
10uM Primer WT F/R	1/1
10uM Primer Neo F/R	1/1
Taq	0.5
DNA	2

Thermal Cycler:

Step 1: 94C, 5min

Step 2: 94C, 30sec

Step 3: 60C, 30sec

Step 4: 72C, 30sec

Step 2 to 4 Cycles: 30

Step 5: 72C, 7min

Taq: Apex and Chromataq 5X Buffer

Bands: MUT: 329bp

WT: 233bp

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

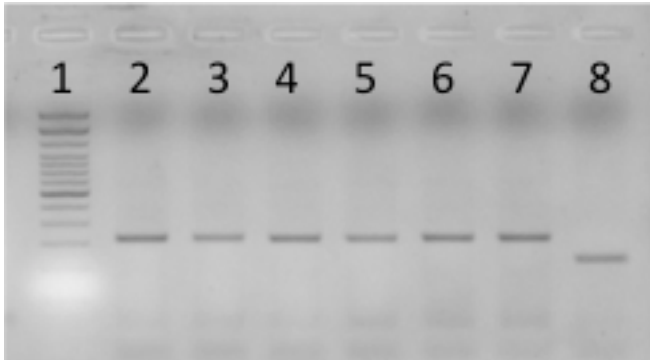
Atp(42051)WTF CTGGCACTGACCACGGCTGAG

Atp(42051)WTR CCAGCCTGTGACTGAGCCCTG

Atp(42051)NeoF CACGACGAGATCCTCGCCGTC

Atp(42051)NeoR GCGCAGCTGTGCTCGACGTTG

Run on 2.0% agarose gel in TAE.



1. 100bp Ladder (Invitrogen)
2. Homozygous Sample
3. Homozygous Sample
4. Homozygous Sample
5. Homozygous Sample
6. Homozygous Sample
7. Homozygous Sample
8. Wild-type Control