

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	34319
MMRRC Strain Name	B6.129S7- <i>Mdm2</i> ^{tm1Ypz} /Mmnc
Gene Name(s)	transformed mouse 3T3 cell double minute 2 (Mdm2)
Breeding Protocol(s)	Backcross to C57BL/6J
Protocol Date	7/1/11

MMRRC #34319 PCR Reaction

There are 2 2-primer reactions. m462-W3& m462WF amplify the wild-type MDM2 allele
 m462-M3& m462MF amplify the MDM2/462 mutant allele

Thermal Cycler:

94°C for 30 sec
 57°C for 45 sec
 72°C for 45 sec
 35x

	1X
DNA	2.0
5X Buffer	5.0
10mM dNTPs	0.5
10 uM Primer F/R	1/1
Taq	0.5
ddH ₂ O	13.0
25mM MgCl ₂	2.0

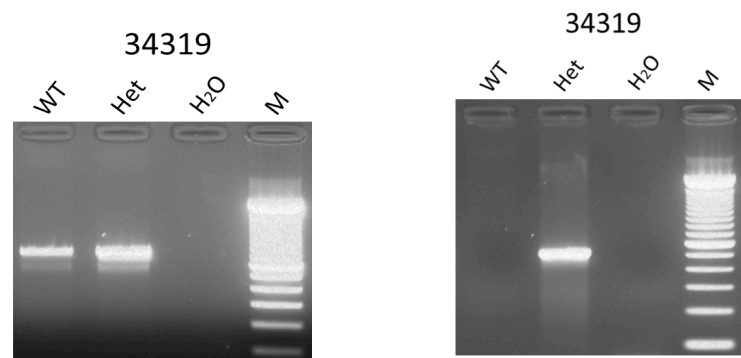
Taq: **Apex and Chromataq 5X Buffer**

Bands: MUTANT: ~520bp
 WT: 714bp

Primer sequences 5' to 3':

m462-M3: 5' – G CTT GTT TCT TTT TTT TAG CTT CTT TGC GGC -3'
 m462-MF: 5'- G AAG GAC TAT TGG AAG TGT ACC TCA TGC-3'
 m462-W3: 5'- GG CTT GTT TCT TTT TTT TAG CTT CTT TGC ACA -3'
 m462WF: 5'- GTA CCT AGA GTC TAC TCT ATT AGC TAA TTT C -3'

Run on 2.0% agarose gel in TAE.



Primers: m462-W3 & m462WF
 M: 100 bp DNA ladder (Invitrogen)

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