

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	31721
MMRRC Strain Name	B6.129S1- <i>Mei1</i> ^{m1Jcs} /Mmnc
Gene Name(s)	Meiosis defective 1 (Mei1)
Breeding Protocol(s)	Backcross to C57BL/6J
Protocol Date	11/11/13

MMRRC# 31721 PCR Reaction

NOTE: Run two, 2-primer reactions for each DNA sample: (M1WT & MOMS-CR) and (M1MMF & MOMS-CR). **Run separately.**

Thermal Cycler

Denaturing: 95C, 30 sec
 Annealing Temp.: 56C, 30 sec.
 Extension: 72C, 30 sec
 Cycles: 35

Taq: **Apex and Qiagen 10X Buffer**

Bands: MUTANT: 227bp (M1MMF reaction)
 WT: 227bp (M1WT reaction)

Primers are 10uM with respect to each primer (20uM total).

Primer sequences 5' to 3':

M1WT: TCACCCAGGCTCTCCTTGCTCAAAA

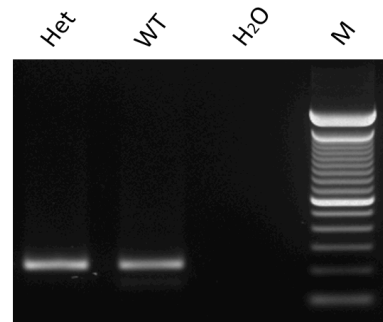
M1MMF: TCACCCAGGCTCTCCTTGCTCAAAT

MOMS-CR: TGTCTCAGGCAGTTGGAGAAGCCCCG

Run on 2.0% agarose gel

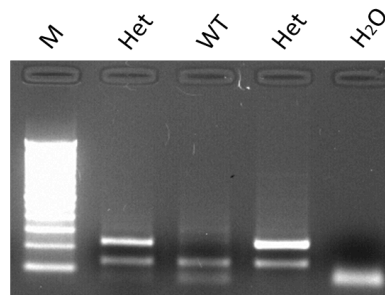
	1X
DNA	1.0
10X Buffer	2.5
10mM dNTPs	1.0
20 uM Primer 1 & 2	1.0
Taq	1.0
ddH ₂ O	16.5
25mM MgCl ₂	2.0

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Primers: M1WT & MOMS-CR
 M: 100 bp DNA ladder (Invitrogen)

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Primers: M1MMF & MOMS-CR
 M: 100 bp DNA ladder (Invitrogen)