

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	31042
MMRRC Strain Name	B6;129- <i>Dnmt3a</i> ^{tm1Jae} /Mmnc
Gene Name(s)	DNA methyltransferase 3A (Dnmt3a)
Breeding Protocol(s)	Sib-Mating
Protocol Date	10/14/13

MMRRC# 31042 PCR Reaction

	1X
DNA	1.0
5X Buffer	5.0
10mM dNTPs	1.0
20 uM Primer 1 & 2	1.0
Taq	1.0
ddH ₂ O	14.5
25mM MgCl ₂	1.5

Denaturing: 95°C for 30 sec
 Annealing Temp.: 60°C for 30 sec
 Extension: 72°C for 30 sec
 Cycles: 32

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

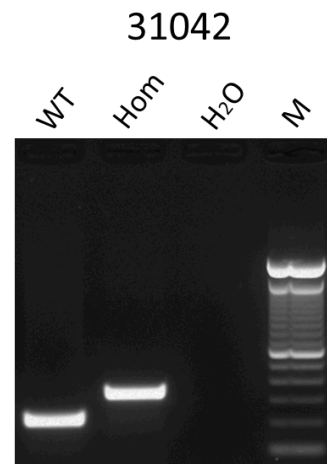
Bands: MUTANT: 250bp
 WT: 150bp

Primer sequences 5' to 3': Primers are 10uM with respect to each primer (20uM total).

3a2LOXfor: CCT CTG GGG AGT TAA GAC TCT TGG CCA GCC C

3a1LOXrev: CCT GTG TGC AGC AGA CAC TTC TTT GGC GTC

Run on 2.5% agarose gel in TAE.



M: 100 bp DNA ladder (Invitrogen)