

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	16820
MMRRC Strain Name	B6;129X1- <i>Hnf1a</i> ^{tm1.1Ylee} /Mmnc
Gene Name(s)	HNF1 homeobox A (Hnf1a)
Breeding Protocol(s)	Sib-mating (-/- usually die before 6 weeks of age)
Protocol Date	2/29/12

MMRRC #16820 PCR Reaction

Thermal Cycler:

Step 1: 94°C for 30 sec
 Step 2: 59°C for 30 sec
 Step 3: 72°C for 45 sec
 35x

	<u>1X</u>
DNA	2.0
5X Buffer	5.0
10mM dNTPs	0.5
10 uM Primer F/R	1/1/1
Taq	0.5
ddH ₂ O	12.0
25mM MgCl ₂	2.0

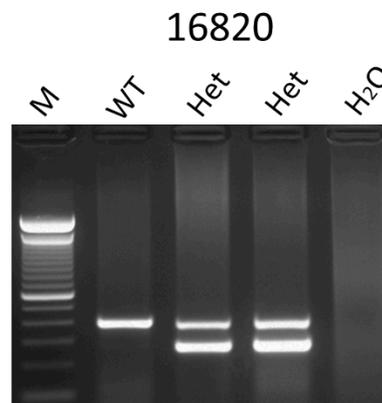
Taq: **Apex and Chromataq 5X Buffer**

Bands: MUTANT: ~280bp
 WT: 405bp, 761bp (usually a fainter band)

Primer sequences 5' to 3':

Hnf1WTF (16820): 5'-CTCGAGTCTGGCCTGAGCAAAGAG-3'
 Common R (16820): 5'-CCTTAACCACCAGCCTGAAGACCA-3'
 Hnf1Mut F (16820): 5'-TGCACTTGCAAGGCTGAAGTCCAA-3'

Run on 2.0% agarose gel in TAE.



M: 100 bp DNA ladder (Invitrogen)