

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	34622
MMRRC Strain Name	B6.129- <i>Isx</i> ^{tm1Shiv} /Mmnc
Gene Name(s)	Intestine specific homeobox (<i>Isx</i>)
Breeding Protocol(s)	Backcross to C57BL/6J
Protocol Date	1/23/14

MMRRC# 34622 PCR Reaction

NOTE: Run 2 reactions for each sample
(WT-F + WT-R for wild-type) and
(WT-F + KO-R for mutant).

Thermal Cycler:

94°C for 30 sec

55°C for 30 sec

72°C for 60 sec

30x

	<u>1X</u>
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: 734bp

WT: 473bp

Primer sequences 5' to 3':

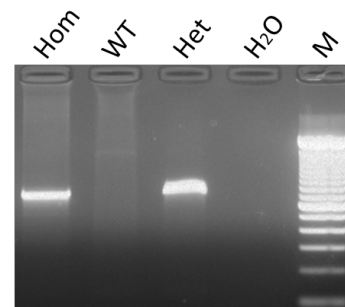
WT-F (34622): TGTTCCCTCAGCTCTCCAC

WT-R (34622): TCAACCCAGCTTCCAGTCTC

KO-R (34622): TCGCCTTCTTGACGAGTTCT

Run on 2.0% agarose gel in TAE.

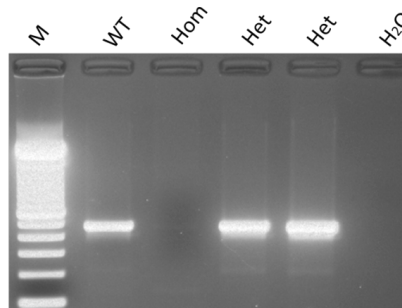
34622



Primers: WT-F + KO-R

M: 100 bp DNA ladder (Invitrogen)

34622



Primers: WT-F + WT-R

M: 100 bp DNA ladder (Invitrogen)

The Mutant Mouse Resource and Research Center – University of North Carolina

CB# 7519, Chapel Hill, NC 27599

Phone: (919)966-4570

www.med.unc.edu/mmrrc

www.mmrrc.org