

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	33006
MMRRC Strain Name	B6.129X1(Cg)- <i>Ptpn4</i> ^{tm1.1Oers} / <i>Mmnc</i>
Gene Name(s)	protein tyrosine phosphatase, non-receptor type 4 (Ptpn4)
Breeding Protocol(s)	Sib-mating
Protocol Date	12/16/13

MMRRC# 33006 PCR Reaction

NOTE: Run 2 reactions for each sample (PtpnF+ PtpnWTR) and (PtpnF+ PtpnMutR).

Thermal Cycler:

94°C for 5 min
 94°C for 45 sec
 59°C for 45 sec
 72°C for 90 sec
 Steps 2-4, 35x
 72°C for 10 min

	1X
DNA	1.0
5X Buffer	5.0
10mM dNTPs	0.5
20 uM Primer F/R	1.0
Taq	1.0
ddH ₂ O	15.0
25mM MgCl ₂	1.5

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

Bands: MUTANT: ~1139bp
 WT: ~660bp

Primer sequences 5' to 3':

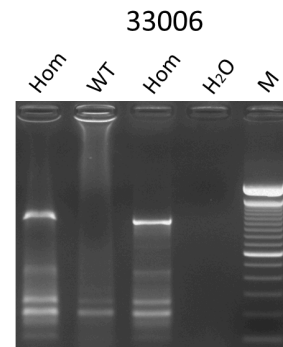
Primers are 10uM with respect to each primer (20uM total).

PtpnF: 5'- CCTGGAATGACAATGTCTTGTGCC

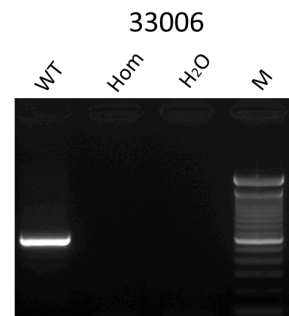
PtpnWTR: 5'- GGTGCAATGTTATCACCATGGAGAG

PtpnMUTR: 5'- GTCCTCGAACTCAGAAATCCACCT

Run on 2.0% agarose gel in TAE.



Primers: PtpnF + PtpnMutR
 M: 100 bp DNA ladder (Invitrogen)



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 M: 100 bp DNA ladder (Invitrogen)