

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	36294
MMRRC Strain Name	129S6.MK- <i>Slc11a2</i> ^{mk/Mdf} /Mmnc
Gene Name(s)	solute carrier family 11 (proton-coupled divalent metal ion transporters), member 2 (Slc11a2)
Breeding Protocol(s)	Sib-mating
Protocol Date	6/27/13

MMRRC #36294 PCR Reaction

Note: 2, 2--primer reactions

Thermal Cycler:

Step 1: 94°C for 5 min

Step 2: 94°C for 30 sec

Step 3: 62°C for 30 sec

Step 4: 72°C for 30 sec

Step 5: Step 2 to 4; Cycles: 30

Step 6: 72°C for 7 min

	<u>1X</u>
ddH ₂ O	13
5X Buffer	5.0
25mM MgCl ₂	2
10mM dNTPs	0.5
10 uM Primer F	1
10 uM Primer R	1
Taq	0.5
DNA	2

Taq: **Apex and chromataq Buffer**

Primers are 10uM with respect to each primer

Primer sequences 5' to 3':

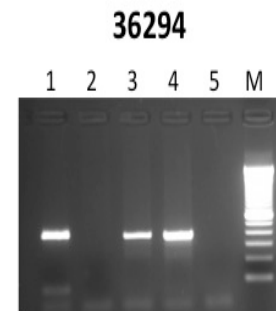
Common-F: CAGCAGACGTAGCATGCATCTTCCA

WT-R: TGTCTGCGATGGTGTGAGGACTGC

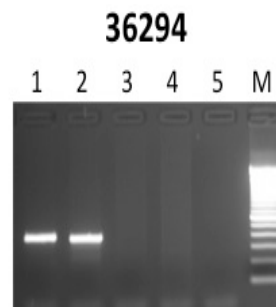
Mut-R: TGTCTGCGATGGTGTGAGGACTGT

Bands: Common-F + WT-R: WT 383 bp
 Common-F + Mut-R: Mut 383 bp

Run on 1% agarose gel in TAE.



Common-F + WT-R (WT). Lane 1: Het; Lane 2: Hom; Lane 3: DI-WT; Lane 4: B6; Lane 5: ddH₂O; M: 100 bp DNA ladder Invitrogen



Common-F + Mut-R. Lane 1: Het; Lane 2: Hom; Lane 3: DI-WT; Lane 4: B6; Lane 5: ddH₂O; M: 100 bp DNA ladder Invitrogen