

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	11888
MMRRC Strain Name	B6.129- <i>Prkci</i> ^{tm1Hed} /Mmnc
Gene Name(s)	protein kinase C, iota (Prkci)
Breeding Protocol(s)	Sib-mating
Protocol Date	10/16/13

MMRRC #11888 PCR Reaction

	1X
ddH ₂ O	12.0
5X Buffer	5.0
25 mM MgCl ₂	2.0
10 mM dNTPs	0.5
10 μM Primer L5	1.0
10 μM Primer L3	1.0
10 μM Primer Neo	1.0
Taq	0.5
DNA	2.0

Thermal Cycler:

Step 1: 94°C for 5 min
 Step 2: 94°C for 30 sec
 Step 3: 58°C for 30 sec
 Step 4: 72°C for 45 sec
 Step 5: 35x from step 2 to step 4
 Step 6: 72°C for 7 min

Taq: Apex and Chromataq 5X Buffer

Primer sequences 5' to 3': Primers are 10 μM with respect to each primer.

L5'endo (11888): AACACCAGGGAGAGTGG

L3'endo (11888): GCCTAGAAAGTAACCCAC

Neo3'S3 (11888): GACGAGTTCTTCTGAGGGG

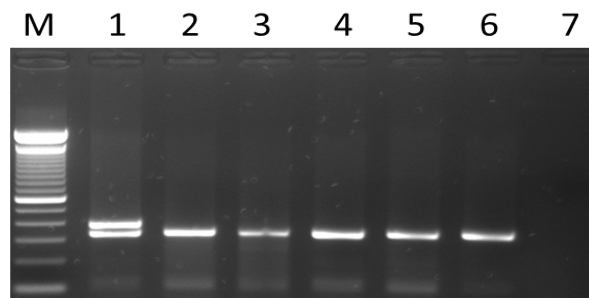
Bands expected:

MUTANT: 400 bp

W: 325 bp

Run on 2.0% agarose gel in TAE.

11888



Lane 1: Het; Lane 2 to 6: WT; Lane 7: H₂O
 M: 100 bp DNA ladder (Invitrogen)