

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	32888
MMRRC Strain Name	B6.FVN-Tg(RP11-12907)9Jwth/Mmnc
Gene Name(s)	phosphoribosyl transferase domain containing 1 (PRTFDC1)
Breeding Protocol(s)	Backcross to C57BL/6NCrI
Protocol Date	7/16/13

MMRRC #32888 PCR Reaction

	<u>1X</u>
ddH ₂ O	13
5X Buffer	5.0
25mM MgCl ₂	2
10mM dNTPs	0.5
10 μM Primer Forward	1.0
10 μM Primer Reverse	1.0
Taq	0.5
DNA	2

Thermal Cycler:

Step 1: 95°C for 5 min

Step 2: 95°C for 30 sec

Step 3: 58°C for 30 sec

Step 4: 72°C for 60 sec

Step 5: 35x from step 2 to step 4

Step 6: 72°C for 10 min

Taq: Apex and Chromataq 5X Buffer

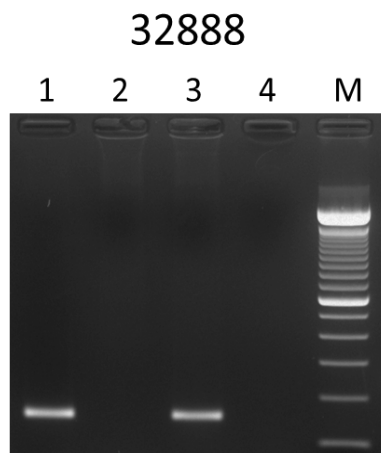
Primer sequences 5' to 3': Primers are 10 μM with respect to each primer.

Intron7.f (32888): CTG AAG TCT GCC TAT GCA AGT G

Intron7.r (32888): TGT TCT CTA GGG CTT CCT CAT C

Bands expected: Tg+ 152 bp

Run on 2% agarose gel in TAE.



Lane 1, 3: Tg+; Lane 2: WT; Lane 4: H₂O; M: 100 bp DNA ladder