

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	42293
MMRRC Strain Name	B6(129S4)- <i>Prdx6</i> ^{tm4.1Abf} /Mmnc
Gene Name(s)	peroxiredoxin 6
Breeding Protocol(s)	Random intra-strain mating
Protocol Date	1/27/2023

PCR Reaction

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

2,2 Primer Reactions (WTF+R for Wild-type and MutF+R for Mutant reaction)

Thermal Cycler:

Step 1: 94C, 5min

Step 2: 94C, 30sec

Step 3: 61C, 30sec

Step 4: 72C, 45sec

Step 2 to 4 Cycles: 30

Step 5: 72C, 7min

Taq: **Apex Taq**

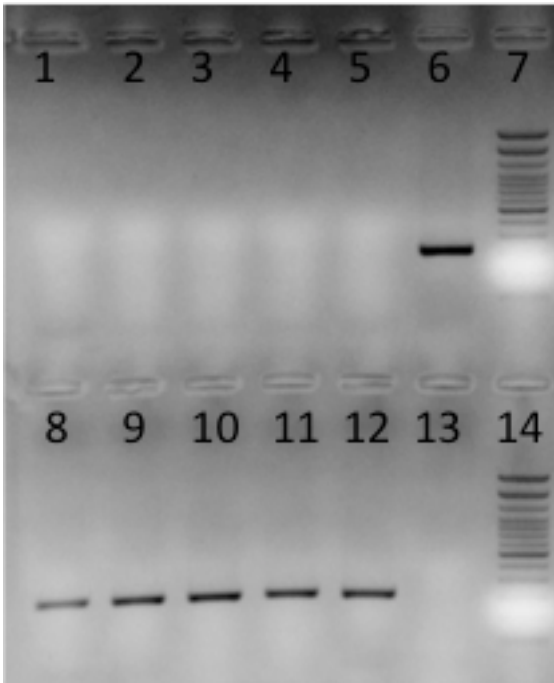
Bands: WT: 317bp MUTANT: 317bp

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

H26A(42293)R: CAGACAACAAACCGTCCTCGGCAAG

H26A(42293)WTF: CATCGGCCGCATCCGCTTCCA

H26A(42293)MutF: CATCGGCCGCATCCGCTTCGC



Wild-type Reaction

1. Homozygous Sample
2. Homozygous Sample
3. Homozygous Sample
4. Homozygous Sample
5. Homozygous Sample
6. Wild-type Control
7. 100bp Ladder (Invitrogen)

Mutant Reaction

8. Homozygous Sample
9. Homozygous Sample
10. Homozygous Sample
11. Homozygous Sample
12. Homozygous Sample
13. Wild-type Control
14. 100bp Ladder (Invitrogen)