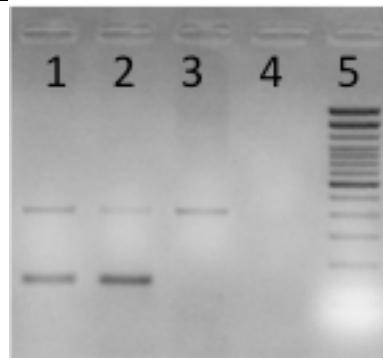


MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	50635
MMRRC Strain Name	B6J.Cg- <i>Rho</i> ^{tm1Phm} /Mmnc
Gene Name(s)	rhodopsin
Breeding Protocol(s)	Sib-mating
Protocol Date	04/29/2020

PCR Reaction

	1X
ddH ₂ O	12
5X Buffer	5.0
25mM MgCl ₂	2
10mM dNTPs	0.5
10uM Primer 1	1
10uM Primer 2	1
10uM Primer 3	1
Taq	0.5
DNA	2



1. Heterozygous Sample
2. Heterozygous Sample
3. Wild-type Control
4. Negative Control
5. 100bp Ladder (Invitrogen)

Thermal Cycler:

Step 1: 94C, 5min
 Step 2: 94C, 30sec
 Step 3: 55C, 30sec
 Step 4: 72C, 45sec
 Step 2 to 4 Cycles: 30
 Step 5: 72C, 7min

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

Bands: WT: 398bp MUTANT: 139bp

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

Rho(50635)F: AGGTTAGAGCTGGAGGACTG
 Rho(50635)R: TAAGACTGATTGGACCATTC
 Rho(50635)Neo: TTCAAGCCCAAGCTTTCGCG