

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	37422
MMRRC Strain Name	B6;129- <i>Stub1</i> ^{tm1Cpat} /Mmnc
Gene Name(s)	STIP1 homology and U-Box containing protein 1 (<i>Stub1</i>)
Breeding Protocol(s)	Intra-strain mating (het x het)
Protocol Date	5/16/18

PCR Reaction

	1X
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

Thermal Cycler:

Step 1: 94°C, 5min
 Step 2: 94°C, 30sec
 Step 3: 58°C, 30sec
 Step 4: 72°C, 30sec
 Step 2 to 4 Cycles: 30
 Step 5: 72°C, 7min

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

Bands: WT: 950bp MUTANT: 290bp

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

Stub1(37422)F: TCA GAG CTA AGG GAA CTG GCT CA

Stub1(37422)R: TCC ATC TCC AGC TGG CAC TGC

Neo1(37422): CTT TTT GTC AAG ACC GAC CTG TCC G

Neo2(37422): CTC GAT GCG ATG TTT CGC TTG GTG