

MMRRC UNC – Genotyping Protocol

MMRRC Strain ID	17226
MMRRC Strain Name	BTNTTF.129S6- <i>Mib2</i> ^{tm1Art} /Mmnc
Gene Name(s)	Mindbomb homolog 2 (Mib2)
Breeding Protocol(s)	Sib-Mating
Protocol Date	10/11/13

MMRRC# 17226 PCR Reaction

	<u>1X</u>
DNA	1.0
10X Buffer	2.5
10mM dNTPs	1.0
10 uM Primer 1 & 2 & 3	1.0
Taq	1.0
ddH ₂ O	17.5
25mM MgCl ₂	1.5

Denaturing: 94C for 45 sec
 Annealing Temp.: 60C for 45 sec
 Extension: 72C for 1min
 Cycles: 40
 Taq: **Apex and Chromataq Buffer**

Primers are 10uM with respect to each primer (30uM total).

Primer sequences 5' to 3':

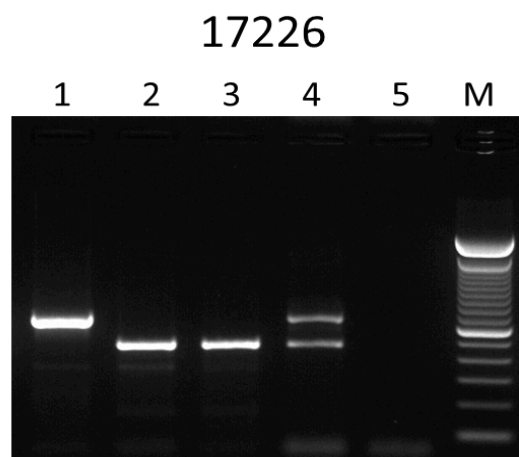
Mib2.7842: 5' GCATCCAGAGCAGGCAAGTTAC

Mib2.8481: 3' ACCAGCAGAGATGGCAGAATGC

Neo66: 5' ATGCTCCAGACTGCCTTG.

Bands: WT: 640bp
 MUTANT: 450bp

Run on 2% agarose gel in TAE.



Lane 1: WT; Lane 2, 3: Hom; Lane 4: Het; Lane 5: H₂O
 M: 100 bp DNA ladder (Invitrogen)