



Welcome to The Genotyping Protocol System

Master Protocol

Strain Name: C57BL/6NJ-Kcnb2^{em1J/J}
Stock Number: 027659
Allele: Kcnb2-6924-KOMP CRISPR
Protocol Name: Kcnb2-6924-KOMP CRISPR
Method: Sanger sequencing
Created: 05-June -2015 (JKELMEN) **Updated:** 05-January -2016 (DTG)

Notes

Mut = X
Notes:
WT= Y

Sequence

Sequence SEQ

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
23901	-	CCT GGC TTG AAC AGA AAG ACA	-	Forward	-
23902	-	CCC GTC CGG TAG AAG TTT AGA	-	Reverse	-

Reaction/Components A

Reaction Components	Volume Amt	Final Concentration	Total Volume Amt
ddH2O	4.55	-	-
5 X Kapa 2G HS buffer	2.40	1	-
25 mM MgCl2	0.96	2	-
10 mM dNTPS-kapa	0.24	.2	-
20 U Primer 1	0.30	.5	-
20 U Primer 2	0.30	.5	-
50 % Glycerol	1.20	5	-
2.5 U/ul Kapa 2G HS taq polymerase	0.05	.01	-
DNA	2.00	-	-

Cycling

Step #	Temp°C	Time	Note
1	94	2 min	-
2	94	20sec	-
3	65	15sec	-0.5 C per cycle decrease
4	68	10sec	-
5	-	-	repeat steps 2-4 for 10 cycles
6	94	15sec	-
7	60	15sec	-
8	72	10sec	-
9	-	-	repeat steps 6-8 for 28 cycles
10	72	2 min	-
11	10	-	hold

Number Of Reactions 1

Version 3.2

