



Welcome to The Genotyping Protocol System

Master Protocol

Strain Name: C57BL/6NJ-*Rab4b*^{em1J}J
Stock Number: 029731
Allele: *Rab4b*^{em1J} Probe
Protocol Name: *Rab4b*^{em1J} Probe
Method: Probe
Created: 30-September-2016 (LUCASM) **Updated:** 10-December -2016 (ESCHAAB)

Notes

Notes: Taqman qPCR protocols are run on a real time PCR instrument. Use an appropriate instrument specific Fluorophore/Quencher combination. The transgene genotype is determined by comparing ΔC_t values of each unknown sample against known homozygous and hemizygous controls, using appropriate endogenous references.

Expected Results: Mutant= 103 bp
Wild Type = 97 bp

Attachments



View [rab4b JW.jpg](#)

- View [Rab4b genomic.gcs](#)
- View [Rab4bem1jMolDesSep2016.docx](#)

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
30003	-	AGC AGG TAC CAT TGA CTA AAG C	-	Common	A
30004	-	CAA TTC TCA CTG CTG GGT TG	-	Wild type Reverse	A
30006	-	CTG GGT TCT CCA TAC CTC ACA	-	Mutant Reverse	A
30007	Hex	CAA AGA CCA TCC ACC AAA GC	Black Hole Quencher 1	WT Probe	-
30008	6-FAM	AAA GCT GCT GAT ATG CTG AGA	Black Hole Quencher 1	MUT Probe	-

Number Of Reactions 1

Reaction A (Endpoint 3 primer)		Cycling (EndPoint)			
Component	Final Concentration	Step #	Temp°C	Time	Note
Kapa Probe Fast QPCR	1 X	1	95	3min	-
ddH2O		2	95	5sec	-
30003	.4 uM	3	60	30sec	-
30006	.4 uM	4	-	-	repeat steps 2-3 for 40 cycles
30004	.4 uM	5	4	-	Forever
Wt Probe	.15 uM				

Reaction A (Endpoint 3 primer)	
Mutant Probe	.15 uM
DNA	

Version 3.2