



Welcome to The Genotyping Protocol System

Master Protocol

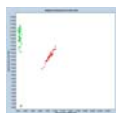
Strain Name: C57BL/6NJ-Nars2^{em1J}/_J
Stock Number: 029628
Allele: Nars2^{em1J}
Protocol Name: Nars2^{em1J}-Alternate 1
Method: Probe
Created: 08-December -2016 (ESCHAAB) **Updated:** 21-December -2016 (JKELMEN)

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= 116 bp
Wild Type = 139 bp

Attachments



View nars2 probe JW.jpg

- View Nars2 genomic.gcs
- View Nars2em1j in progress 8-29-16.docx

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
29444	-	CTC AAC TTC CCT AAT TGT GTG TC	-	Wild type Forward	A
29445	-	GGC AGA GGA TAC CAG CAT GT	-	Common	A
29446	-	CAA TCT AAT GCT CTC TCT GGC TGA C	-	Mutant Forward	A
29448	Hex	CTT TGA AGA AAA AGG AAC CAG GAG	Black Hole Quencher 1	WT Probe	-
30965	6-FAM	AGA TTG TGA ATA CCT CTA ACA TTT TCA C	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	-
29444	.4 uM	3	60	30sec	-
29445	.4 uM	4	-	-	repeat steps 2-3 for 40 cycles
29446	.4 uM	5	4	-	Forever
Wt Probe	.15 uM				

Reaction A (Endpoint 3 primer)

Mutant Probe	.15 uM
DNA	

Version 3.2