



Welcome to The Genotyping Protocol System

Master Protocol

Strain Name: C57BL/6NJ-Ap4e1^{em1J}/J
Stock Number: 026839
Allele: Ap4e1^{em1J}
Protocol Name: Ap4e1^{em1J}
Method: End Point Analysis
Created: 17-July -2015 (JKELMEN) **Updated:** 15-March -2016 (LUCASM)

Notes

Notes: This allele from project Ap4e1_6638-F-2877 was generated at The Jackson Laboratory by injecting Cas9 RNA and 3 guide sequences, GCAATCAAGTTGGCCCAACA, TGTCCTGACTGTCGTTTTTA and CTGTTTGTTCATAGTGAT which resulted in a 1bp insertion **A** in exon 3 beginning at Chromosome 2 positive strand position 127014235bp (GRCm38). This mutation is predicted to cause amino acid sequence changes after residue 105 and early truncation 19 amino acids later.

Expected Results: Mutant = A/A
Heterozygote = -/A
Wild type = -/-

Attachments



[View](#) 26839.JPG

- [View](#) Ap4e1_em1j__6-30-15[1].docx
- [View](#) Ap4e1 genomic.gcs

Sequence

Sequence TGGTTATATACATGCAATCAAGTTGGCCCA**A**CAAGGAAACCTCTTAGAAAAAAGA
GTGG

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
24321	-	CGA TGA AGG AAT GTA TGG TGA	-	Forward	-
24322	-	TCA GGA CAG AAG TCT TAA ACA TAT ACA	-	Reverse	-
24323	Hex	TTG GCC CAA CAA GGA AAC	Black Hole Quencher 1	WT Probe	-
24324	6-FAM	TTG GCC CAA ACA AGG AAA C	Black Hole Quencher 1	MUT Probe	-

Reaction/Components A

Reaction Components	Volume Amt	Final Concentration	Total Volume Amt
2 X Kapa Probe Fast QPCR	2.50	.42	-

Cycling

Step #	Temp°C	Time	Note
1	95	10min	-
2	95	15sec	-

Reaction/Components A				Cycling			
ddH2O	7.44	-	-	3	60	1min	repeat steps 2-3 for 40 cycles
100 uM 24321	0.02	.17	-				
100 uM 24322	0.02	.17	-				
100 uM 24323	0.01	.08	-				
100 uM 24324	0.01	.08	-				
DNA	2.00	-	-				

Number Of Reactions 1

Version 3.2