C57BL/6J-*Tbk1*^{em3Lutzy}/1J

Stock No: 027080
Protocol 22051: End Point Analysis Assay - Tbk1<em#3(G217R)Lutzy>
Version 2.0

Notes

The genotyping protocol(s) presented here have been optimized for reagents and conditions used by The Jackson Laboratory (JAX). To genotype animals, JAX recommends researchers validate the assay independently upon receipt of animals into their facility. Reaction cycling temperature and times may require additional optimization based on the specific genotyping reagents used.

Expected Results

Mutant = A/A
Heterozygote = G/A
Wild type = G/G

Sequence

CTTCAGCATCCGGACATGTATGAAAGGGCAGTGCTAAGA
AAGGACCATCAGAAGAAGTACGG
GGCTACCGTTGATCTGTGGAGTGTTGGAGTGACATTCTAC
CATGCAGCCACG(g/a)GGTCGCTG
CCGTTTAGACCCTTCGAGGGGCCTCGGAGGAACAAAGAA
GTAATGTAAGCACCT

JAX Protocol

Protocol Primers

PRIMER	5' LABEL	SEQUENCE 5' → 3'	3' LABEL	PRIMER TYPE	REACTION	NOTE
24482		TGT GGA GTG TTG GAG TGA CA		Forward	A	
24483		GGT GCT TAC ATT ACT TCT TTG TTC C		Reverse	A	
24484	Fluorophore-1	AGC CAC GGG GTC GCT	Quencher-1	WT Probe		
24488	Fluorophore-2	CAG CCA CGA GGT CGC TG	Quencher-2	MUT Probe		

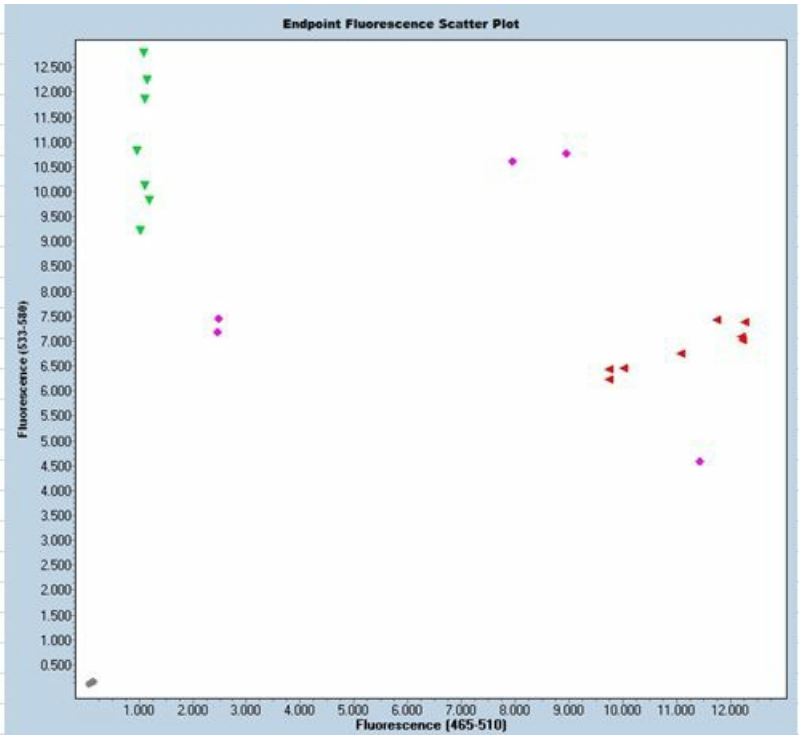
Reaction A

COMPONENT	FINAL CONCENTRATION
Kapa Probe Fast QPCR	1.00 X
ddH2O	
24482	0.40 uM
24483	0.40 uM
Wt Probe	0.15 uM
Mutant Probe	0.15 uM
DNA	

Cycling

STEP	TEMP °C	TIME	NOTE
1	95.0	--	
2	95.0	--	
3	60.0	--	
4		--	repeat steps 2-3 for 40 cycles
5	4.0	--	Forever

JAX uses a very high speed Taq (~1000 bp/sec), use cycling times recommended for your reagents.


Endpoint Fluorescence Scatter Plot
Fluorescence (533-580)
Fluorescence (465-510)
12.500
12.000
11.500
11.000
10.500
10.000
9.500
9.000
8.500
8.000
7.500
7.000
6.500
6.000
5.500
5.000
4.500
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