



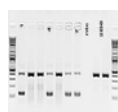
Welcome to The Genotyping Protocol System

Master Protocol

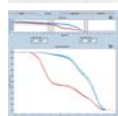
Strain Name: C57BL/6NJ-*Paf1^{em1J}*
Stock Number: 029329
Allele: *Paf1^{em1J}*
Protocol Name: *Paf1^{em1J}*
Method: MELT
Created: 15-June -2016 (JKELMEN) **Updated:** 10-August -2016 (ESCHAAB)

Notes

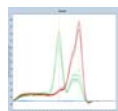
Expected Results: Mutant = 104 bp
 Heterozygote = 104 bp and 400 bp
 Wild type = 400 bp

Attachments

View 29329 Paf1 gel.jpg



View 29329 Paf1 hrn.JPG



View 29329 Paf1 mca.jpg

- View Paf1em1j_in progress 6-10-16.docx
- View Paf1 genomic.gcc

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
28498	-	GGC ACA TAG GAG GTG CTC AG	-	Common	A
28500	-	GTA AGG GGC AGG AGG ACC A	-	Mutant Reverse	A

Number Of Reactions 1

Reaction A (2 Primer)		Cycling (TouchDown 65-60_12-12-11)			
Component	Final Concentration	Step #	Temp°C	Time	Note
ddH2O		1	94	2 min	-
Kapa 2G HS buffer	1.3 X	2	94	20sec	-
MgCl2	2.6 mM	3	65	15sec	-0.5 C per cycle decrease
dNTPS-kapa	.26 mM	4	68	10sec	-
28498	.5 uM	5	-	-	repeat steps 2-4 for 10 cycles (Touchdown)
28500	.5 uM	6	94	15sec	-
		7	60	15sec	-

Reaction A (2 Primer)		Cycling (TouchDown 65-60_12-12-11)			
Glycerol	6.5 %	8	72	10sec	-
Dye	1 X	9	-	-	repeat steps 6-8 for 28 cycles
Kapa 2G HS taq polymerase	.03 U/ul	10	72	2 min	-
DNA		11	10	-	hold

Version 3.2