



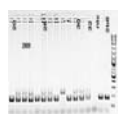
Welcome to The Genotyping Protocol System

Master Protocol

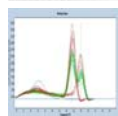
Strain Name: C57BL/6NJ-*Rusc1*^{em1J/J}
Stock Number: 029009
Allele: *Rusc1*^{em1J}
Protocol Name: *Rusc1*^{em1J}
Method: MELT
Created: 04-April -2016 (JKELMEN) **Updated:** 21-July -2016 (ESCHAAB)

Notes

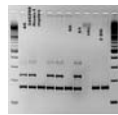
Expected Results: Mutant = 229 bp
 Heterozygote = 162 bp and 229 bp
 Wild type = 162 bp

Attachments

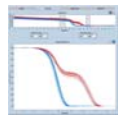
View 29009.jpg



View 29009 tm.JPG



View 29009 Rusc1 gel.jpg



View 29009 Rusc1 hrm.JPG

View Rusc1em1j in progress 3-28-16.docx

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
27461	-	AGA AGC CCA GAG TGG GAC TG	-	Common	A
27462	-	TAT CAG GAG ACC TGG GTG GA	-	Wild type Reverse	A
27463	-	TCC AGT GAT GAG GTC CTT CC	-	Mutant Reverse	A

Number Of Reactions 1

Reaction A (3 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
		4	68	10sec	-

Reaction A (3 primer)		Cycling (TouchDown 65-60_12-12-11)			
dNTP KAPA	.26 mM	5	-	-	repeat steps 2-4 for 10 cycles (Touchdown)
27463	.5 uM	6	94	15sec	-
27462	.5 uM	7	60	15sec	-
27461	.5 uM	8	72	10sec	-
Glycerol	6.5 %	9	-	-	repeat steps 6-8 for 28 cycles
Dye	1 X	10	72	2 min	-
Kapa 2G HS taq polymerase	.03 U/ul	11	10	-	hold
DNA					

Version 3.2