



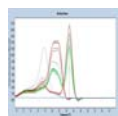
Welcome to The Genotyping Protocol System

**Master Protocol**

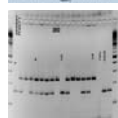
**Strain Name:** C57BL/6NJ-*Rab11fip3<sup>em1J</sup>*  
**Stock Number:** 028992  
**Allele:** *Rab11fip3<sup>em1J</sup>*  
**Protocol Name:** *Rab11fip3<sup>em1J</sup>*  
**Method:** MELT  
**Created:** 05-April -2016 (JKELMEN) **Updated:** 20-July -2016 (ESCHAAB)

**Notes**

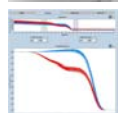
Expected Results: Mutant = 239 bp  
 Heterozygote = 153 bp and 239 bp  
 Wild type = 153 bp

**Attachments**

View Rab11-28992 Melt.JPG



View Rab11-28992 Gel.jpg



View 28992 Rab11fip3 hrm.JPG

- View Rab11fip3em1jin progress 3-25-16.docx

- View Rab11fip3 genomic1.gcs

**Protocol Primers**

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
27529	-	TTG GTT CCA TAT CCT CTC TGC	-	Common	A
27530	-	TGC TGG TGT CCT CAT CTG TG	-	Wild type Reverse	A
27531	-	TCA TCC TTG AAC ACA GAG CAA G	-	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
dNTP KAPA	.26 mM	4	68	10sec	-
27531	.5 uM	5	-	-	repeat steps 2-4 for 10 cycles (Touchdown)
		6	94	15sec	-

<b>Reaction A (3 primer)</b>		<b>Cycling (TouchDown 65-60_12-12-11)</b>			
27529	.5 uM	7	60	15sec	-
27530	.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