



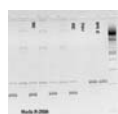
Welcome to The Genotyping Protocol System

Master Protocol

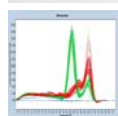
Strain Name: C57BL/6NJ-*Muc3a*^{em1J/J}
Stock Number: 029384
Allele: *Muc3a*^{em1J}
Protocol Name: *Muc3a*^{em1J} Alternate1
Method: MELT
Created: 21-October -2016 (LUCASM) **Updated:** 28-October -2016 (ESCHAAB)

Notes

Expected Results: Mutant = 161 bp
 Heterozygote = 161 bp and 215 bp
 Wild type = 215 bp

Attachments

View 29384 muc3a.jpg



View muc3a MCA JW.jpg

- View Muc3a genomic.gcs
 - View Muc3aem1j in progress 6-24-16.docx

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
30312	-	GCT TGC TGC TCT AGC ATG G	-	Common	A
30313	-	CAG AAG AGC GGT CCA ACC	-	Wild type Forward	A
30314	-	AGA GAG CTA CAC CCT AGC ACC T	-	Mutant Forward	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	-
30312	.5 uM	5	-	-	repeat steps 2-4 for 10 cycles (Touchdown)
30314	.5 uM	6	94	15sec	-
30313	.5 uM	7	60	15sec	-
Glycerol	6.5 %	8	72	10sec	-
		9	-	-	repeat steps 6-8 for 28 cycles

Reaction A (3 primer)		Cycling (TouchDown 65-60_12-12-11)			
Dye	1 X	10	72	2 min	-
Kapa 2G HS taq polymerase	.03 U/ul	11	10	-	hold
DNA					

Version 3.2