



Welcome to The Genotyping Protocol System

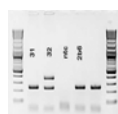
Master Protocol

Strain Name: C57BL/6NJ-*lqca*^{em1J/J}
Stock Number: 029136
Allele: *lqca*^{em1J}
Protocol Name: *lqca*^{em1J}
Method: MELT
Created: 09-May -2016 (LUCASM) **Updated:** 04-August -2016 (ESCHAAB)

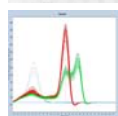
Notes

Expected Results: Mutant = 344 bp
 Heterozygote = 226 bp and 344 bp
 Wild type = 226 bp

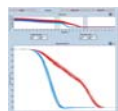
Attachments



View 29136 lqca gel.jpg



View 29136 lqca mca.jpg



View 29136 lqca hrn.JPG

- View *lqca* Genomic.gcs
- View *lqcaem1j*MolDesApr2016.docx

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
27958	-	AGT GTG CCC AGT GGA TAG CA	-	Common	A
27959	-	AGA CAT TCT CCA TGT TCC TGA	-	Wild type Reverse	A
27960	-	CAG CTG TAA CCT ACT GGG TCA 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	-
MgCl ₂	2.6 mM	3	65	15sec	-0.5 C per cycle decrease
dNTP KAPA	.26 mM	4	68	10sec	-
27959	.5 uM	5	-	-	repeat steps 2-4 for 10 cycles (Touchdown)
		6	94	15sec	-

Reaction A (3 primer)		Cycling (TouchDown 65-60_12-12-11)			
27960	.5 uM	7	60	15sec	-
27958	.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