



Welcome to The Genotyping Protocol System

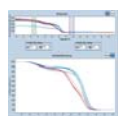
Master Protocol

Strain Name: C57BL/6NJ-Zfp560^{em1J}
Stock Number: 029279
Allele: Zfp560^{em1J}
Protocol Name: Zfp560^{em1J}-alternate1
Method: High Resolution Melting
Created: 14-October -2016 (JKELMEN) **Updated:** 21-October -2016 (ESCHAAB)

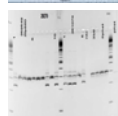
Notes

Expected Results: Mutant = 237 bp
 Heterozygote = 237 bp and 299 bp
 Wild type = 299 bp

Attachments



View Zfp HRM.JPG



View Zfp560-29279 Gel.jpg

- View Zfp560em1jMolDesMay2016-1.docx
- View Zfp560 genomic.gcs

Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
30242	-	GAG AGA GAG AGC ACT TGA TTT CC	-	Mutant Forward	A
30243	-	TGA AAT GAT GTT CAC AAT AAT CG	-	Common	A
30244	-	GAT GGA AAA GAC GGT GTC TGA	-	Wild type 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	-
30244	.5 uM	5	-	-	repeat steps 2-4 for 10 cycles (Touchdown)
30242	.5 uM	6	94	15sec	-
30243	.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