



## Welcome to The Genotyping Protocol System

## Master Protocol

**Strain Name:** C57BL/6NJ-*Fastkd5<sup>em1</sup>J/J*  
**Stock Number:** 026913  
**Allele:** *Fastkd5<sup>em1</sup>J*  
**Protocol Name:** *Fastkd5<sup>em1</sup>J*  
**Method:** Sanger sequencing  
**Created:** 30-December -2014 (JKELMEN) **Updated:** 28-January -2015 (JKELMEN)

## Sequence

Sequence SEQ

## Protocol Primers

Primer	5' Label	Sequence 5' --> 3'	3' Label	Description	Reaction
22338	-	TCT TGG CAT AGA TCC TGT TGC	-	Forward	-
22339	-	CGG CTG ACA TTC TGG AGT CT	-	Reverse	-

## Reaction/Components A

Reaction Components	Volume Amt	Final Concentration	Total Volume Amt
ddH2O	42.55	-	-
5 X Kapa 2G HS buffer	2.40	.24	-
25 mM MgCl <sub>2</sub>	0.96	.48	-
10 mM dNTPS-kapa	0.24	.05	-
20 U Primer 1	0.30	.12	-
20 U Primer 2	0.30	.12	-
50 % Glycerol	1.20	1.2	-
2.5 U/ul Kapa 2G HS taq polymerase	0.05	0	-
DNA	2.00	-	-

## Cycling

Step #	Temp °C	Time	Note
1	94	2 min	-
2	94	20sec	-
3	65	15sec	-0.5 C per cycle decrease
4	68	10sec	-
5	-	-	repeat steps 2-4 for 10 cycles
6	94	15sec	-
7	60	15sec	-
8	72	10sec	-
9	-	-	repeat steps 6-8 for 28 cycles
10	72	2 min	-
11	10	-	hold

Number Of Reactions 1

Version 3.2