

**GENOTYPING BY PCR PROTOCOL  
MUTANT MOUSE REGIONAL RESOURCE CENTER: UC DAVIS**

[mmrrc@ucdavis.edu](mailto:mmrrc@ucdavis.edu)

530-754-MMRRC

Please provide the following information required for genetic analysis of your mutant mice.

*Note to MAC users:* to ensure your graphic can be viewed on a PC please follow the steps below when inserting the graphic into this document. DO NOT drag and drop or copy/paste the graphic into this document.

- Open the original graphic in the program that created it
- Choose File, Save As
- Select No Compression in the save options.
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- Switch to Word, choose Insert, Picture, From File and choose the newly saved picture.

*These instructions are very generic. The menu options for your graphics program may be different.*

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Strain Name		MMRRC Stock Number
<b>1318-CreER-IRES-GFP</b>		<b>037164</b>

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NAME OF PCR: 1318-CreER-IRES-GFP

MMRRC: 037164-UCD

Protocol: *(PCR protocol provided by Donating Investigator)*

Reagent/Constituent	Volume (µL)
Water	17.75
10x Buffer (contains 15mM MgCl <sub>2</sub> )	2.5
MgCl <sub>2</sub> (stock concentration is mM)	0
Betaine (stock concentration is 5M) <i>Optional</i>	0
dNTPs (stock concentration is 10mM)	1.5
Qiagen Q Buffer	0
Primer 1. (stock concentration is 20µM) Name: <b>1318.for</b>	0.5
Primer 2. (stock concentration is 20µM) Name: <b>1318.rev</b>	0.5
Primer 3. (stock concentration is 20µM)	0
Primer 4. (stock concentration is 20µM)	0
Taq Polymerase 5Units/µL	0.25
DNA (example) extracted w/ "Qiagen DNeasy columns or other similar silica based kits"	2
<i>The total volume is auto-calculated based on volumes entered, right click the total and update field to show/recalculate the total volume.</i>	<b>TOTAL VOLUME OF REACTION: 25 µL</b>

Comments on protocol:

Strategy:

Steps	Temp (°C)	Time (m:ss)	# of Cycles
1. Initiation/Melting <span style="float:right">HOT START? <input type="checkbox"/></span>	95	5min	1
2. Denaturation	95	30sec	
3. Annealing <span style="float:right">steps 2-3-4 cycle in sequence</span>	58	30sec	39x
4. Elongation	72	30sec	
5. Amplification	72	10min	1
6. Finish	4	∞	n/a

Primers:

Electrophoresis Protocol:

Name	Nucleotide Sequence (5' - 3')	Argarose: 1	V:120
1.		Estimated Running:Time: 45 min.	
2. <b>1318.for</b>	GGTTCAAAGCTGAGAAGCGCC	Primer Combination	Band
3. <b>1318.rev</b>	ACACTATCCCGTCTTGGGAGA	#2+#3	500 bp
4.			N/A bp
5.			bp
			Genotype
			KO or MT/-
			WT +/-

*Please size gel images to fit in this space*

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**Protocol / Gel Comments:**

