

**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
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*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>953-CreER-IRES-GFP</b>		<b>37571</b>

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NAME OF PCR: 953-CreER-IRES-GFP

MMRRC: 37571

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

Reagent/Constituent	Volume (μL)
<b>2x Buffer</b> – GoTaq Green Master Mix (Promega) Contains:	6
MgCl <sub>2</sub> 3mM	
Proprietary compound that increases sample density	
dNTPs 400 μM	
Yellow and Blue Dyes	
Bacterially derived Taq DNA Polymerase	
<b>Primer Mix</b> (1.25 μM of each primer in water)	5
Primer 1 Name: 953.F	
Primer 2. Name: CreER.R	
DNA From Tail Digest (2mm tail digested with proteinase K and diluted to a final volume of 200 ul)	1
<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:</b>
	12 μL

Comments on protocol:

Strategy:

Steps	Temp (°C )	Time (m:ss)	# of Cycles
1. Initiation/Melting HOT START? <input type="checkbox"/>	94	5min	1
2. Denaturation	94	1 min	
3. Annealing steps 2-3-4 cycle in sequence	58	1 min	34x
4. Elongation	72	1 min	
5. Amplification	72	10min	1
6. Finish	16	∞	n/a

Primers:

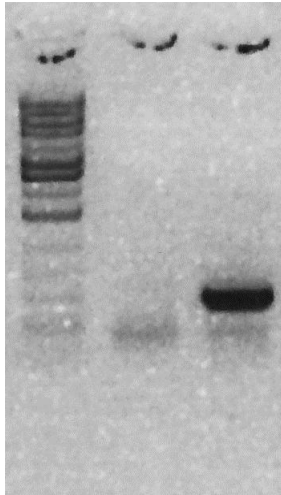
Electrophoresis Protocol:

Name	Nucleotide Sequence (5' - 3')	Agarose: 1% V:120		
1.		Estimated Running:Time: 30 min.		
2. 953.F	GCG AAG TCA GCA CCA ACA TA	<b>Primer Combination</b>	<b>Band</b>	<b>Genotype</b>
3. CreER.R	AGTGCTGCCTCTGACCTCAT	953.F/ CreER.R	486 bp	KO or MT/-
4.		953.F/ CreER.R	No Band	WT +/-
5.				

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**Protocol / Gel Comments:**



Lane 1 = All Star 1kb DNA Ladder  
Lane 2 = Tail DNA from negative (wild-Type) mouse  
Lane 3 = Tail DNA from positive 953CT2IG mouse

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