

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

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530-754-MMRRC

NAME OF PCR: STOCK Tg(SOX2-cre/ERT2, GFP)1040Jlr MMRRC: 037134-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
dNTPs (stock concentration is 10mM)	1.5
Primer 1. (stock concentration is 20μM)	0.5
Primer 2. (stock concentration is 20μM)	0.5
Taq Polymerase 5Units/μL	0.25
DNA extracted with <input type="checkbox"/> NaOH <input type="checkbox"/> Proteinase K <input type="checkbox"/> Other:	2.0
<b>TOTAL VOLUME OF REACTION:</b>	<b>25.000 μL</b>

Comments on protocol:

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Strategy:

Steps	Temp (°C)	Time (m:ss)	# of Cycles
1. Initiation/Melting <span style="float:right">HOT START? <input type="checkbox"/></span>	94	5:00	1
2. Denaturation	94	0:30	
3. Annealing <span style="float:right">steps 2-3-4 cycle in sequence</span>	57	0:30	<b>39x</b>
4. Elongation	72	0:30	
5. Amplification	72	10:00	1
6. Finish	4	∞	n/a

Primers:

Name	Nucleotide Sequence (5' - 3')	Argarose: 1% V: 120
1. 192.for	TATTGCTGTGCTTTGGGTGA	Estimated Running Time: <b>45</b> min.
2. 192.rev	CACCTGCAAATGGGAAAAGT	
		<b>Primer Combination</b>
		1 and 2
		<b>Band (bp)</b>
		500
		<b>Genotype</b>
		Knockout

