

**GENOTYPING BY PCR PROTOCOL
MUTANT MOUSE REGIONAL RESOURCE CENTER**

sacoord@mmrrc.org

800-910-2291 North America, +1-530-757-5710 International

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.
- Save as JPG or PNG or similar format that's compatible with both PC and Mac Word versions.
- 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.

Donating Investigator/PI		
John C. Crabbe, PhD		
Email		
crabbe@ohsu.edu		
Institution		
VA Portland Health Care System		
Address		
3710 SW US Veterans Hospital Road, R&D-12		
City	State	Zip
Portland	OR	97239
Lab Contact		
Dr. Pamela Metten		
Email		
mettenp@ohsu.edu		
Telephone	FAX	
503-220-8262 x56680	503-721-1029	
Strain Name		MMRRC Stock Number
Withdrawal Seizure Control, Replicate 1		

**GENOTYPING BY PCR PROTOCOL
MUTANT MOUSE REGIONAL RESOURCE CENTER**

sacoord@mmrrc.org

800-910-2291 North America, +1-530-757-5710 International

NAME OF PCR: Not applicable, this is a selected line **MMRRC:** 0-CTR

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

Reagent/Constituent	Volume (µL)
Water	
10x Buffer	
MgCl ₂ (stock concentration is mM)	
Betaine (stock concentration is 5M) <i>Optional</i>	
dNTPs (stock concentration is 10mM)	
DMSO <i>Optional</i>	
Primer 1. (stock concentration is 20µM)	
Primer 2. (stock concentration is 20µM)	
Primer 3. (stock concentration is 20µM)	
Primer 4. (stock concentration is 20µM)	
Taq Polymerase 5Units/µL	
DNA (50-200ng/ µL) extracted w/ "Qiagen DNeasy columns or other similar silica based kits"	
<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>	TOTAL VOLUME OF REACTION:
	0.000 µL

Comments on protocol:

•

Strategy:

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

Primers:

Electrophoresis Protocol:

Name	Nucleotide Sequence (5' - 3')	Agarose:	V:
1.		Estimated Running:Time: min.	
2.		Primer Combination	Band
3.			bp
4.			bp
5.			bp

Please size gel images to fit in this space

**GENOTYPING BY PCR PROTOCOL
MUTANT MOUSE REGIONAL RESOURCE CENTER**

sacoord@mmrrc.org

800-910-2291 North America, +1-530-757-5710 International

Protocol / Gel Comments:

Gel pictures: