

RMRC Genotyping Protocol-PCR

No.	RMRC13408	Strain Name	C57BL/6- <i>Uap111</i> ^{em2Yen} /Narl
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A. Primer

	Primer name	Sequence
1	Uap111-short Fw	5'-TTGAGGCCGGAAGAGTTAGAG -3'
2	Uap111-short Rev	5'-TCCAGCTCAATATGGCAAGTC-3'

B. Genomic extraction :

- Phenol-Chloroform extraction commercial extraction kit _____
 Lysis buffer _____ other _____

C. Reaction Conditions: for Uap111 wt allele, Uap111 flox allele

Component	Concentration		Volume		Step	Temperature, °C	Time, min:sec	Number of Cycles
H ₂ O			9.5	μl	1	95	5:00	1
Ampliqon Taq DNA Polymerase 2x Master Mix RED	2X		12.5	μl	2	95	00:30	30
						60	00:40	
						72	00:30	
Primer 1	Uap111-short Fw	10 μM	1	μl	3	72	05:00	
Primer 2	Uap111-short Rev	10 μM	1		4	4	pause	
DNA	--		1	μl	5			
Total			25	μl	6			

The information of Taq, including company name and catalog number :

Taq DNA Polymerase 2x Master Mix RED, 2 mM MgCl₂; Ampliqon #A190303

D. PCR Product

	Product size		Target region
1	Uap111-wt	623 bp	
2	Uap111-flox	703 bp	

E. Gel Photo

