

RMRC Genotyping Protocol-PCR

No.	RMRC13430	Strain Name	C57BL/6-Tg(Oog1,-cre; CAGGS-flEmGFP-TyrmIR)19Narl/Narl
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A. Primer

	Primer name	Sequence
1	Cre313F	CCACGACCAAGTGACAGCAATG
2	Cre685B	CAGAGACGGAAATCCATCGCTC
3	b-actin F	GAACATGGCATTGTTACCAACTGG
4	b-actin B2	AAGGGAGTCCTATGGGAGAACG

B. Genomic extraction :

- ☐ Phenol-Chloroform extraction ☐ commercial extraction kit _____
- ☒ Lysis buffer Allele-In-One ABP-PP-MT01500 Mouse Tail Direct Lysis Buffer ☐ other _____

C. Reaction Conditions

Step 1. PCR

Component	Concentration		Volume		Step	Tempera- ture, °C	Time, sec	Number of Cycles
H ₂ O	--		8	μl	1	95	300	1
PCR buffer	10x		-	μl	2	95	30	35
PCR Dye Marker Mix	2x		10	μl	3	60	30	
Primer 1	10	μM(ρmol/μl)	0.4	μl	4	72	30	
Primer 2	10	μM(ρmol/μl)	0.4	μl	5	Go to step 2		
Primer 3	10	μM(ρmol/μl)	0.4					
Primer 4	10	μM(ρmol/μl)	0.4					
DNA	50~250ng/ml		0.4	μl	6	72	300	1
Total			20	μl				

The information of Taq, including company name and catalog number : Taq DNA Polymerase 2x

Master Mix RED (Ampliqon, Cat.No.:A180303)

PCR Product

	Primer set		Product size	Target region
1	Cre313F	Cre685B	373 bp	Cre
2	b-actin F	b-actin B2	245 bp	b-actin

D. Gel Photo

