

RMRC Genotyping Protocol-PCR

No.	RMRC13385	Strain Name	C57BL/6-Tg (Miox-Nanoluc)6Narl/Narl
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A. Primer

	Primer name	Sequence
1	Miox E15F	5'- CAT TAA CTT GCT GAG GTC AGG AGG -3'
2	N-Luc 5B	5'- CGT CCG AAA TAG TCG ATC ATG TTC -3'
3	Miox E13B	5'- TTA AGA GGC AGT GAT CTC CAC CTG -3'

B. Genomic extraction :

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

C. Reaction Conditions

Reaction Conditions								
Component	Concentration		Volume		Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--		40	μl	1	95	2min	1
PCR buffer	10x		5	μl	2	95	30sec	35
dNTP	10	mM	1	μl	3	60	30sec	
Primer 1	20	μM(ρmol/μl)	1	μl	4	72	45sec	
Primer 2	20	μM(ρmol/μl)	0.5	μl	5	72	10min	1
Primer 3	20	μM(ρmol/μl)	0.5	μl				
gDNA	50~250 ng/ul		1	μl				
Taq	5	U/μl	1	μl				
Total			50	μl				

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

D. PCR Product

	Primer set		Product size	Target region
1	Miox E15F	N-Luc 5B	514bp	
2	Miox E15F	Miox E13B	449bp	

E. Gel Photo

