

RMRC Genotyping Protocol-PCR

No.	RMRC13398	Strain Name	C57BL/6- <i>Lama3^{em1Tmh}</i> /Narl
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A. Primer

	Primer name	Sequence
1	5F	5'-TGGTGGGTCAGGACCATCCC- 3'
2	3B	5'-AGGTCAAGTTGGGAAAAGAAGACC- 3

B. Genomic extraction :

- ☐ Phenol-Chloroform extraction
 ☐ commercial extraction kit _____

☐ Lysis buffer _____
 ☐ other _____

C. Reaction Conditions

Component	Concentration	Volume	Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--	μl	1			
PCR buffer	10x	μl	2			
dNTP	mM	μl	3			
Primer 1	μM(ρmol/μl)	μl	4			
Primer 2	μM(ρmol/μl)	μl	5			
DNA	--	μl	6			
Taq	U/μl	μl	7			
Mg ²⁺	mM	μl	8			
Total		μl	9			

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

D. PCR Product

	Primer set		Product size	Target region
1	5F	3B	262 bp	Wild type and Knock-in allele: 262 bp Knockout allele < 262 bp
2	BtsCI		bp	Wild type: 170 + 92 bp Knockin : 262 bp

E. Gel Photo

電泳圖：

