

RMRC Genotyping Protocol-PCR

No.	RMRC13399	Strain Name	C57BL/6N-Tg(BGLAP-Mir29a)Wfs/NarI
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A. Primer

	Primer name	Sequence
1	BamHI-pre_mir-29a-F	5'gag gat ccc ctc aag gat acc aag gga tga at 3'
2	XbaI-pre_mir-29a-R	5'ctt cta gaa gga gtg ttt cta ggt atc cgt ca 3'

B. Genomic extraction :

■ commercial extraction kit illustra tissue and cell genomicpre mini spin kit

☐ Lysis buffer _____ ☐ other _____

C. Reaction Conditions

Component	Concentration		Volume		Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--		37.6	μl	1	94	5m	
PCR buffer	10x		5	μl	2	94	10s	
dNTP	2.5	mM	2	μl	3	60	20s	
Primer 1	10	μM(μmol/μl)	1	μl	4	72	35s	
Primer 2	10	μM(μmol/μl)	1	μl	5	72	7m	
DNA	--		1	μl	6			
Taq	2	U/μl	0.4	μl	7			
Mg ²⁺	1.5	mM	0	μl	8			
Total			50	μl	9			

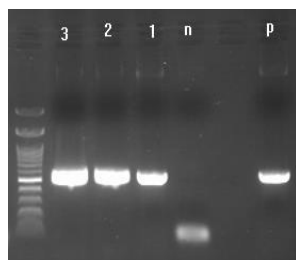
The information of Taq, including company name and catalog number :

D. PCR Product

	Primer set	Product size	Target region
1		506 bp	

E. Gel Photo

1. 1pg construct
2. 2pg construct
3. 3pg construct
4. only wild type



5. Positive