

RMRC Genotyping Protocol-PCR

No.	RMRC13178	Strain Name	C57BL/6-Tg(Tnnt2-EGFP-Mir28)LB03IBMS/CCC
-----	-----------	-------------	--

A. Primer

	Primer name	Sequence
1	Tnt-EGFP junction-F	5' CTGAGGCTGAGCAGACACCTC
2	Tnt-EGFP junction-R	5' CCGGTGGTGCAGATGAACTTC

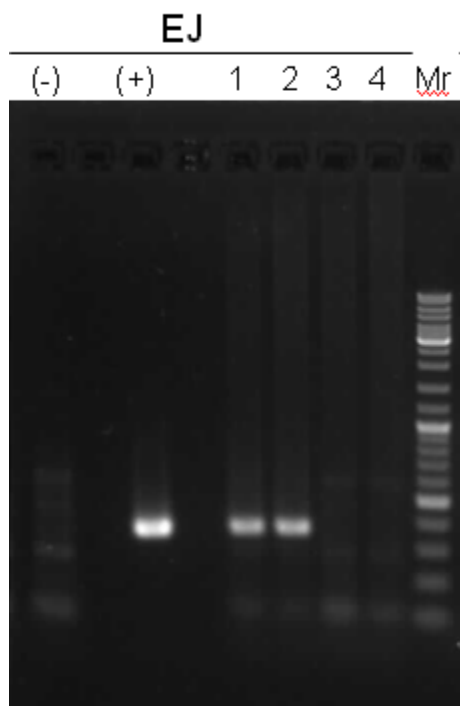
B. Reaction Conditions

Component	Concentration		Volume		Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--		3.6	μl	1	94	300	
PCR buffer	10x			μl	2	94	15	30
dNTP		mM		μl	3	58	30	30
Primer 1	10	μM(μmol/μl)	0.2	μl	4	72	30	30
Primer 2	10	μM(μmol/μl)	0.2	μl	5	72	600	
DNA	crude tail lysate		1	μl	6	4	--	
Taq	Tiangen 2x Taq mastermix (blue)		5	μl	7			
Mg ²⁺		mM		μl	8			
Total			10	μl	9			

C. PCR Product

	Primer set		Product size	Target region
1	Tnt-EGFP junction-F	Tnt-EGFP junction-R	359 bp	

Gel Photo



(-) Ctrl: B6 WT, (+) Ctrl: Tg-miR-28/28* construct , 1~4: Tg

mice tail samples =>Result: No.1, 2 are Tg-rcTnT-miR-28/28*; No. 3,4 are WT littermates