

RMRC Genotyping Protocol-PCR

No.	RMRC13219	Strain Name	C57BL/6-Tg(pCAGGS-flpe)15Narl
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A. Primers

No.	Primer name	Sequence
1	FlpF	CAGCAATCAAGAGAGCCACATTC
2	FlpR	CGTTTTAGGACTGGTTCAGAGTTCC
3	Il2F	CTAGGCCACAGAATTGAAAGATCT
4	Il2R	GATGGTGGAAATTCTAGCATCATCC

B. Reaction Conditions

Component	Concentration	Volume	Step	Temperature, °C	Time, sec	Number of cycles
H ₂ O	—	10.2 μ l	1	94	180	1
PCR buffer	10 x	1.5 μ l	2	94	30	35
Formamide	—	0.5 μ l	3	60	30	35
dNTP	10 mM	0.3 μ l	4	72	30	35
Primer 1	20 μ M (μ mol/ μ l)	0.3 μ l	5	72	180	1
Primer 2	20 μ M (μ mol/ μ l)	0.3 μ l				
Primer 3	10 μ M (μ mol/ μ l)	0.3 μ l				
Primer 4	10 μ M (μ mol/ μ l)	0.3 μ l				
gDNA	50-250 ng/ μ l	1 μ l				
Taq	5 U/ μ l	0.3 μ l				
Total		15 μ l				

C. PCR Product

No.	Primer set		Product size	Target region
1	FlpF	FlpR	625 bp	Flp
2	Il2F	Il2R	324 bp	Wildtype

D. Gel Photo

