RMRC Genotyping Protocol-PCR

No.	RMRC13231	Strain Name	C57BL/6-Tg(pCAGGS-Flpe)14Narl/Narl

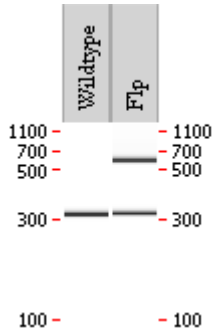
A. Primers

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_2O	–	10.2 µl	1	94	180	1
PCR buffer	10 x	1.5 µl	2	94	30	35
Formamide	–	0.5 µl	3	60	30	35
dNTP	10 mM	0.3 µl	4	72	30	35
Primer 1	20 µM (pmol/µl)	0.3 µl	5	72	180	1
Primer 2	20 µM (pmol/µl)	0.3 µl				
Primer 3	10 µM (pmol/µl)	0.3 µl				
Primer 4	10 µM (pmol/µl)	0.3 µl				
gDNA	50-250 ηg/µl	1 µl				
Taq	5 U/µl	0.3 µl				
Total		15 µl				

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