

RMRC Genotyping Protocol-PCR

No.	RMRC13142	Strain Name	B6.129-Grid1<tm2b(LacZ)Vda>
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A. Primer

	Primer name	Sequence
1	For437	CCATGCTGATCTCTTCCTGTC
2	Rev438	GGTCGCTACAGACGTTGTTTG
3	Rev440	GAGCATGACATCATTGAGACG

B. Reaction Conditions

Component	Concentration		Volume		Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--		19	μl	1	94	180	1
PCR buffer	10x		2.5	μl	2	94	30	30
dNTP		mM		μl	3	56	30	30
Primer 1	10	μM(μmol/μl)	1	μl	4	68	120	30
Primer 2	10	μM(μmol/μl)	1	μl	5	68	600	1
DNA	--		1	μl	6			
Taq	AccuPrime	U/μl	0.5	μl	7			
Mg ²⁺		mM		μl	8			
Total			25	μl	9			

C. PCR Product

	Primer set		Product size	Target region
1	For437	Rev438	363 bp	GFP
2	For437	Rev440	266 bp	

D. Gel Photo

