

RMRC Genotyping Protocol-PCR

No.	RMRC13101	Strain Name	B6.129- <i>Gata1</i> ^{tm225(S26A)} Jyen
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A. Primer

	Primer name	Sequence
1	GATA1-Ex2 FS	5'-ACT GGC CTA CTA CAG AGA AGC TGA G-3'
2	GATA1-Outside LoxP	5'- AAG ACA GCT TCG TAC ACA CTT CCA G -3'

B. Reaction Conditions

Component	Concentration		Volume		Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--		16.1	μl	1	94		
PCR buffer	10x		2.5	μl	2	94 65 72	30 60 30	5
dNTP	2.5	mM	2	μl	3	94 63 72	30 60 30	5
Primer 1	10	μM(μmol/μl)	0.5	μl	4	94 61 72	30 60 30	5
Primer 2	10	μM(μmol/μl)	0.5	μl	5	94 57 72	30 60 30	20
DNA	--		2	μl	6	72	120	1
Taq	5	U/μl	0.2	μl	7	4	∞	1
DMSO			1.25	μl	8			
Total			25	μl	9			

C. PCR Product

	Primer set		Product size	Target region
1	1	2	250 bp	Wild type
2	1	2	370 bp	mutant

D. Gel Photo

GATA1-primer

