

RMRC Genotyping Protocol-PCR

No.	RMRC13376	Strain Name	C57BL/6- <i>Cyp11a1</i> ^{tm10Bcc} /Narl
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A. Primer

	Primer name	Sequence
1	mSCC1_347F	5'-ATTGGTGCCCTGACTACTGG-3'
2	mSCC1_617R	5'-GGCAGGCTTTCTTGTCATTC-3'

B. Reaction Conditions

Component	Concentration	Volume	Step	Temperature, °C	Time, sec	Number of Cycles
H ₂ O	--	7.5	μl	1	95	300
2x Dream Taq PCR Master Mix	2x	10	μl	2	95	30
			μl	3	56	60
Primer 1	10	μM(ρmol/μl)	1	μl	4	72
Primer 2	10	μM(ρmol/μl)	1	μl	5	72
DNA	--	0.5	μl	6		
			μl	7		
			μl	8		
Total		20	μl	9		

The brand (廠牌) and product name (品名) of Taq :

2x Dream Taq PCR Master Mix #K1071 (Thermo Scientific)

C. PCR Product

	Primer set	Product size	Target region
1	mSCC1_347F / mSCC1_617R	271 bp (WT) / 372 bp (MUTANT)	Intron I, <i>Mus musculus Cyp11a1</i> , ENSMUSG00000032323

D. Gel Photo

100bp F/F F/F WT F/+ t

