

RBRC 11075

Strain name C57BL/6N-Ttc5<em1Rbrc>

Primer	primer_name	primer_seq	length	Reference
1	exon5-intron5	CTCAAGCAGTAAGTACTTGCAATCACC	27	RIKEN BRC
2	intron 5	ACATGGCTTACAACATGCGAGTTCC	25	RIKEN BRC
3	intron 2	GAGCTCCTGTAAGTGGGTGGCA	22	RIKEN BRC

Reaction Components

Enzyme TOYOBO KOD FX (Code.No : KFX-101)

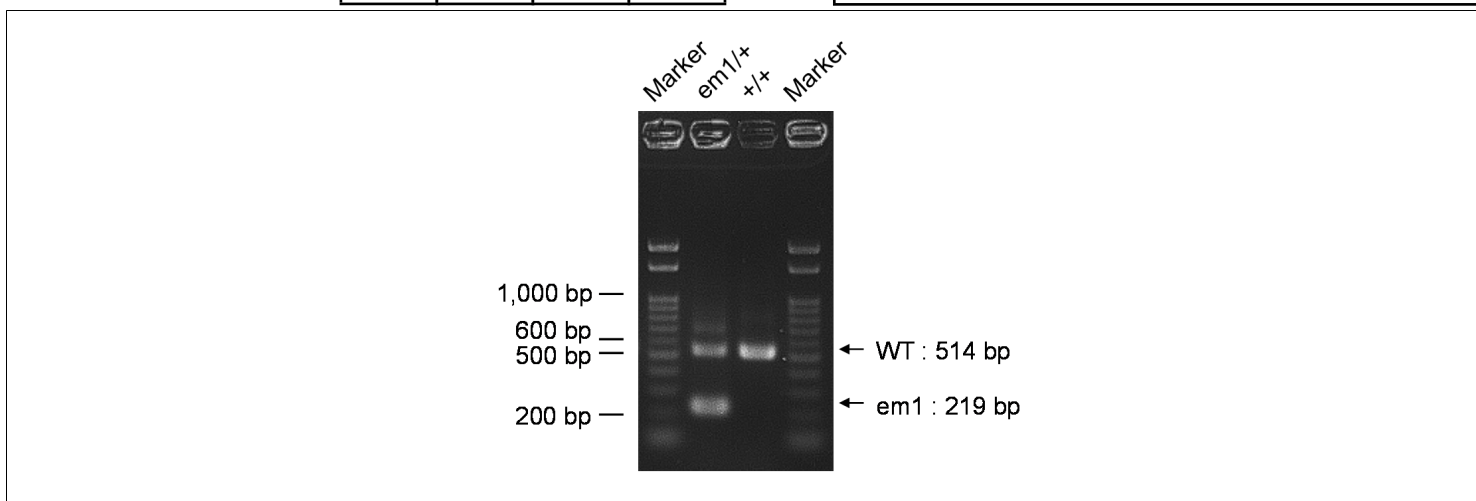
Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30*	
4	68	60*	*Repeat for 30 cycles

Reagent	Conc	Reaction 1	2	3	4
H2O		1.8			
PCR buffer	2 X	10.0			
dNTP	2.0 mM	4.0			
primer 1	10 pmol/μl	1.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
DNA		1.0			
Taq	1 U/μl	0.2			
total		20			

(μL)

Marker	Gene Ladder 100 (Wako, Cat. 316-06951)
Gel	2% agarose in 1 X TAE (100 V, 30 min)



primer_set	product_size	product (allele type)	
primer 1	primer 2	514 bp	WT
primer 2	primer 3	219 bp	em1

+/+ (wild-type): 514 bp
em1/+ (heterozygous): 514 bp and 219 bp

Comment Standard PCR

Novagen KOD Xtreme™ Hot Start DNA Polymerase can be purchased from EMD.