PCR protocol

Sheet ID PS_08200
Release date : 9/16/2022
Print date : 9/16/2022

RBRC 11387 **Strain name** C57BL/6N-Pmepa1<em1Akt>

Primer	primer_name	primer_seq	length		Reference
1	WT_exon 2	GCTGGAGTTCGTGCAAATCGTGGTCGT	27		RIKEN BRC
2	mut_exon 2	GCTGGAGTTCGTGCAAATCGTGGTCGA	27		RIKEN BRC
3	intron 2	CTGTCAGAGTGTTCCGTGCGGAGG	24		RIKEN BRC

Reaction Components

Enzyme QIAGEN Multiplex PCR Kit (Cat. No. 206143)

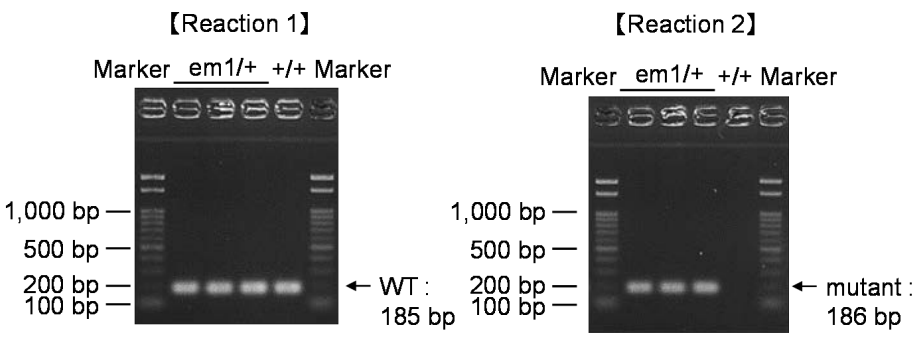
Reagent	Conc	Reaction 1	2	3	4	
H2O		2.0	2.0			(μL)
Master Mix	2 X	5.0	5.0			
primer 1	10 pmol/μl	1.0	0			
primer 2	10 pmol/μl	0	1.0			
primer 3	10 pmol/μl	1.0	1.0			
DNA		1.0	1.0			
	total	10	10			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	95	900	
2	94	30	*
3	60	90	*
4	72	60	* *Repeat for 30 cycles

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 3	185 bp	WT
primer 2	primer 3	186 bp	mutant

Reaction 1
+/+ : 185 bp
em1/+ : 185 bp
Reaction 2
+/+ : not detected
em1/+ : 186 bp

Comment