PCR protocol

Sheet ID PS_07863
Release date : 11/10/2021
Print date : 11/10/2021

RBRC 10586 **Strain name** B6.Cg-Chil3<em1(Venus)Mtka>

Primer	primer_name	primer_seq	length		Reference
1	recF	GATTATTCGCAAACACTTCC	20		Depositor's primer
2	recR	TGCTTCCTTGTTAGATCCTG	20		Depositor's primer
3	Venus-R	TTCTGCTGGTAGTGGTCGGC	20		Sci. Immunol., 3(28)(2018).

Reaction Components

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

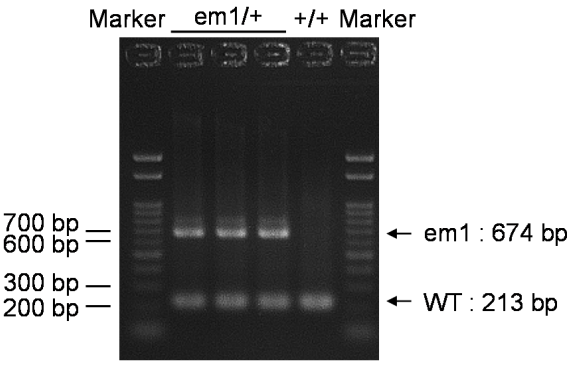
Reagent	Conc	Reaction 1	2	3	4	
H2O		1.8				(μL)
PCR buffer	2 X	10.0				
dNTP	2.0 mM	4.0				
primer 1	10 pmol/μl	1.0				
primer 2	10 pmol/μl	1.0				
primer 3	10 pmol/μl	1.0				
DNA		1.0				
Taq	1 U/μl	0.2				
	total	20				

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10	*
3	55	30	*
4	68	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 2	213 bp	WT	+/+ (wild-type): 213 bp em1/+ (heterozygous): 213 bp and 674 bp
primer 1	primer 3	674 bp	em1	

Comment Standard PCR

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