PCR protocol

Sheet ID PS_08186
Release date : 6/10/2021
Print date : 6/10/2021

RBRC 10736

Strain name C57BL/6J-Tg(Cacna1i-cre)NIto

Primer	primer_name	primer_seq	length		Reference
1	intron 1	GCACCGAGACCTATGCACACAC	22		RIKEN BRC
2	exon 2	CTGCTGAAGATGAGGGTGGTAAG	23		RIKEN BRC
3	CW-Cre2	TCCGGTTATTCAACTTGCACCATGC	25		Nature, 406(6797): 726-731 (2000).

Reaction Components

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

Reagent	Conc	Reaction 1	2	3	4
H2O		1.8			
PCR buffer	2 X	10.0			
dNTP	2 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			

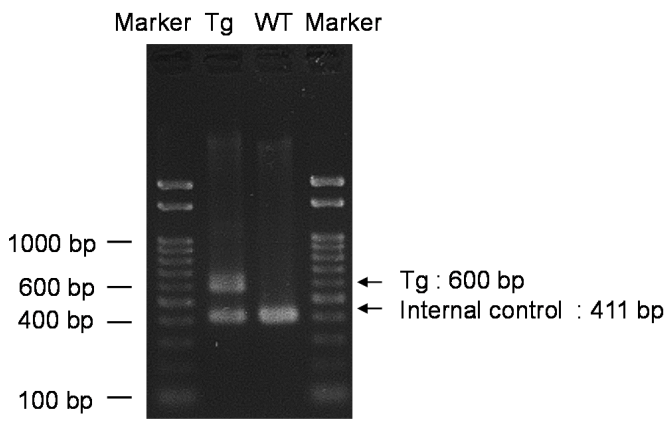
(μL)

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10 *	
3	60	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	411 bp	Internal control
primer 1	primer 3	600 bp	Tg

Non-Tg : 411 bp
Tg : 411 bp, 600 bp

Comment Standard PCR

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