PCR protocol

Sheet ID PS_08321
Release date : 5/6/2022
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RBRC 11609 **Strain name** B6D2-1700026J12Rik<em1Nino>

Primer	primer_name	primer_seq	length		Reference
1	1700026J12Rik-F	ACCTGTATGTTCCAACACAGCT	22		Depositor's primer
2	common	CTCAATTCTGTCACTTCACATGTG	24		RIKEN BRC
3	wild	CTCCAACATAGAGGCACAAGAC	22		RIKEN BRC
4	1700026J12Rik-KOR	ATAATTCATTGATGATGAGA	20		Depositor's primer

Reaction Components

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

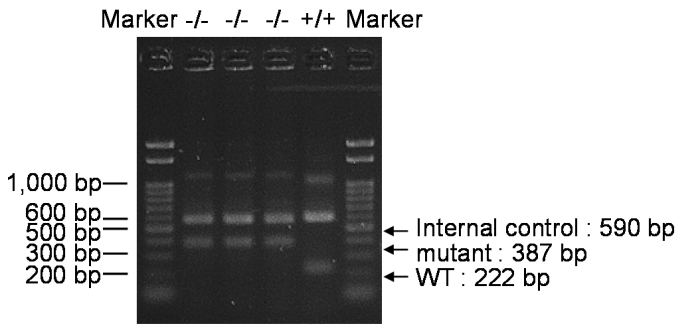
Reagent	Conc	Reaction 1	2	3	4	
primer 1	10 pmol/μl	1.0				(μL)
primer 2	10 pmol/μl	1.0				
primer 3	10 pmol/μl	1.0				
primer 4	10 pmol/μl	1.0				
Master Mix	2 X	5.0				
DNA		1.0				
	total	10				

Reaction Conditions

step	temp (℃)	time (sec)	note
1	95	900	
2	94	30	*
3	55	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 2	590 bp	Internal control	+/+ (wild-type): 590 bp and 222 bp -/- (homozygous): 590 bp and 387 bp
primer 2	primer 3	222 bp	WT	
primer 1	primer 4	387 bp	mutant (em1)	

Comment Separated PCR