PCR protocol

Sheet ID PS_06110
Release date : 1/5/2018
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RBRC 06459 **Strain name** C57BL/6J-Tyr<em2Utr>

Primer	primer_name	primer_seq	length		
1	wild	CCCCTTCAAAGGGGTGGATGACAG	24		RIKEN BRC
2	mut	CCCCTTCAAAGGGGTGGATGACAT	24		RIKEN BRC
3		GCTTCATGGGCAAAATCAAT	20		Depositor's primer

Reaction Components

Enzyme

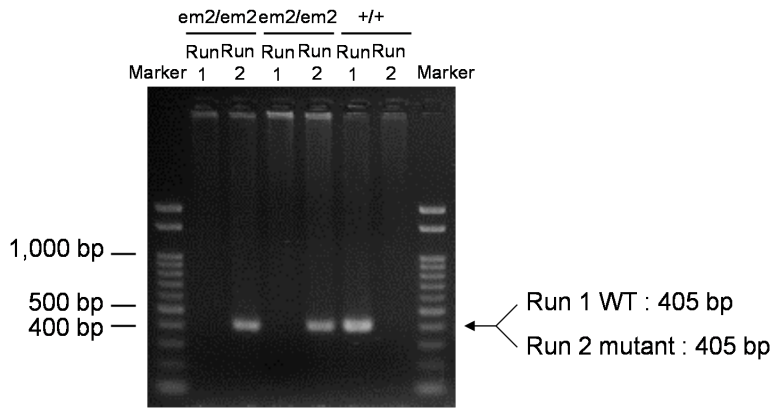
QIAGEN Multiplex PCR Kit (Cat. No. 206143)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.0	2.0		
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
5	72	600		

Allele specific PCR (ASP)



Marker

Gene Ladder 100 (Wako, Cat. 316-06951)

Gel

2% agarose in 1 X TAE (100 V, 30 min)

PCR products	primer_set	product_size	product
primer 1	primer 3	405 bp	WT
primer 2	primer 3	405 bp	mutant

Comment