PCR protocol

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RBRC 09877 Strain name C57BL/6N-Bclaf1<em1Nuc>

Primer	primer_name	primer_seq	length		
1	Btf_2ndcoding for	CCGACAACCACTGAGGATCCGTGCCTACTTTCCCAGG	38		Depositor's primer
2	WT	GGTCGTCTCATTCCTCTATTATTCCTGTAATC	32		RIKEN BRC
3	MT_em1	GGTCGTCTCATTCCTCTATTATTCCTGTATC	31		RIKEN BRC

Reaction Components

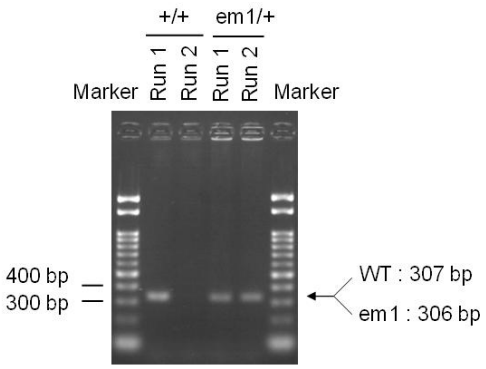
Enzyme: QIAGEN Multiplex PCR Kit (Cat. No. 206143)

	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	1.0		
primer 2	10 pmol/μl	1.0	0		
primer 3	10 pmol/μl	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	65	90	*
4	72	60	* *Repeat for 25 cycles

Separated PCR



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 2	307 bp	WT
primer 1	primer 3	306 bp	em1

Comment