PCR protocol

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RBRC 09375 Strain name B6.D2-(D1Mit191-D1Mit14)/Hkv

Primer	primer_name	primer_seq	length		
1	D1Mit191-1	AGGCATGCCAGTTTCCTTC	19		Depositor's primer
2	D1Mit191-2	GGGAAATATTTTCCCTTTTTGG	22		Depositor's primer
3	D1Mit14_1	GCCAGACAGGGCTACATTGT	20		Depositor's primer
4	D1Mit14_2	AGACTGAACTCTGGCCTCCA	20		Depositor's primer

Reaction Components

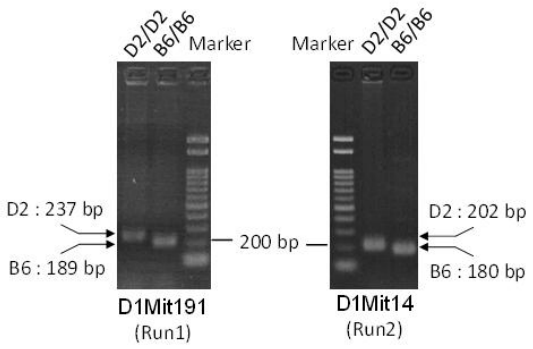
Enzyme: QIAGEN Multiplex PCR Kit (Cat. No. 206143)

	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	1.0	0		
primer 3	10 pmol/μl	0	1.0		
primer 4	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 30 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	237 bp	D2
primer 1	primer 2	189 bp	B6
primer 3	primer 4	202 bp	D2
primer 3	primer 4	180 bp	B6

Comment: WHT<BALB