PCR protocol

Sheet ID PS_03858
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RBRC 04698 Strain name A/J-7A<SM>

Primer	primer_name	primer_seq	length		
1	D7Mit95-1	GACAGACAAATGACAATAACTGCC	24		Depositor's primer
2	D7Mit95-2	AGAACTCAGGTGGAAATTTTGG	22		Depositor's primer
3	D7Mit355-1	AGTAAGGAAAATGGAAATACCAACA	25		Depositor's primer
4	D7Mit355-2	GATACCACAGAATTTTTGGATGC	23		Depositor's primer
5	D7Mit17-1	CTGGCATTTATGTTGCTTCA	20		Depositor's primer
6	D7Mit17-2	AACTTGCCTTCTGTCCTCCA	20		Depositor's primer

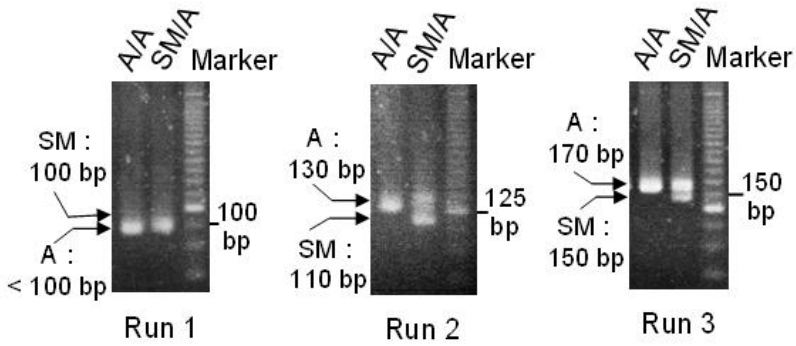
Reaction Components

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

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.0	2.0	2.0	
Master Mix	2 X	5.0	5.0	5.0	
primer 1	10 pmol/μl	1.0	0	0	
primer 2	10 pmol/μl	1.0	0	0	
primer 3	10 pmol/μl	0	1.0	0	
primer 4	10 pmol/μl	0	1.0	0	
primer 5	10 pmol/μl	0	0	1.0	
primer 6	10 pmol/μl	0	0	1.0	
DNA		1.0	1.0	1.0	
	total	10	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	60	1800		



Marker

25 bp DNA ladder (Invitrogen, Cat. 10597-011)

Gel

4% Nusieve in 0.5 X TBE (100 V, 60 min)

PCR products

primer_set		product_size	product
primer 1	primer 2	< SM, 100 bp	A, SM
primer 3	primer 4	130, 110 bp	A, SM
primer 5	primer 6	170, 150 bp	A, SM

Comment D7Mit95: 5% Nusieve gel in 0.5 X TBE (100 V, 120 min)