PCR protocol

RBRC 03817 **Strain name** B6D2F1-Polr3a<Gt(LVtrap1)LG04Osb>

Primer	primer_name	primer_seq	length		
1	4163	GTCACTCAATCCTCTGCCTTTG	22		Depositor's primer
2	4582	TTTCACACAAGTGACTCTGTGTTGG	25		Depositor's primer
3	4135	GATCTCTAGTTACCAGAGTCA	21		Depositor's primer

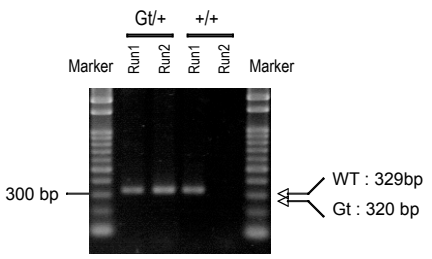
Reaction Components

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		13.3	13.3		
PCR buffer	10 X	2.0	2.0		
dNTP	2.5 mM	1.6	1.6		
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		
Taq	5U/μl	0.1	0.1		
	total	20	20		

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30*	
3	60	30*	
4	72	60*	*Repeat for 30 cycles
5	72	300	



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	329 bp	WT
primer 1	primer 3	320 bp	Gt

Comment