PCR protocol

Sheet ID PS_02539
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RBRC 03154 **Strain name** Pcdhg-DgHS4-5(49)

Primer	primer_name	primer_seq	length		
1	gHS4-5 T-C F	AGGAATTGTAAAAGAATCATGAAGTCCTAGGC	32		Depositor's primer
2	gHS4-5 wild R	TCATGGTTCTGGGTGTGATGG	21		Depositor's primer
3	gHS4-5 T-F R2	TGCCAAGAGATGTTTGCTCTGGC	23		Depositor's primer

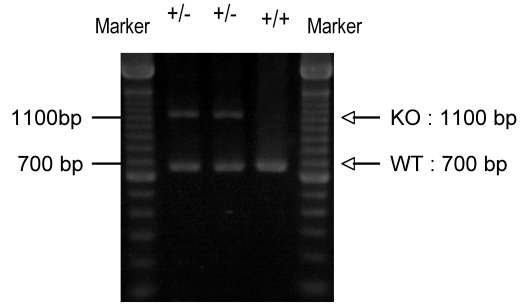
Reaction Components

Enzyme TaKaRa LA Taq with GC buffer (Cat. No. RR02AG)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.8			
PCR buffer	2 X GC I	10.0			
dNTP	2.5 mM	2.0			
primer 1	10 pmol/μl	2.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
DNA		1.0			
Taq		0.2			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30	*
3	57	30	*
4	72	60	* *Repeat for 30 cycles
5	72	300	



Marker

100 bp ladder (Invitrogen, Cat. 15628-050)

Gel

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

PCR products

primer_set		product_size	product
primer 1	primer 2	700 bp	WT
primer 1	primer 3	1100 bp	KO

Comment