PCR protocol

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RBRC 05168 **Strain name** C57BL/6-Tg(Th-FLPo)4Imayo

Primer	primer_name	primer_seq	length		
1	FW_Primer	CCTGTGACAGTGGATGCAATT	21		RIKEN BRC
2	FLPo-287bp-rv	CCTCGCTGCTCTCGAACTGCAGCTG	25		Depositor's primer
3	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24		JAX Genotyping Protocol
4	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25		JAX Genotyping Protocol

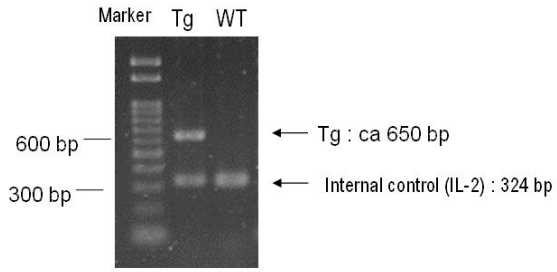
Reaction Components

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		11.3			
PCR buffer	10 X	2.0			
dNTP	2.5 mM	1.6			
primer 1	10 pmol/μl	1.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
primer 4	10 pmol/μl	1.0			
DNA		1.0			
Taq	5 U/μl	0.1			
	total	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	ca 650 bp	Tg
primer 3	primer 4	324bp	Internal control (IL-2)

Comment