PCR protocol

RBRC 02731 **Strain name** C57BL/6J-Tg(Dsg3TCR140)1

Primer	primer_name	primer_seq	length		
1		GTCTCCCGGGTGCACTCAAGGACCAAGTGT	30		Depositor's primer
2		GCCCGCGGCCTGCCAGGAAGTCTAGTCAAAA	31		Depositor's primer
3		GTCTCGAGTTTCTCTTTTAACTAATGCCC	29		Depositor's primer
4		GCCCGCGGCGTAGGACTGTGAACAGTACAT	30		Depositor's primer
5	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24		JAX Genotyping Protocol
6	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25		JAX Genotyping Protocol

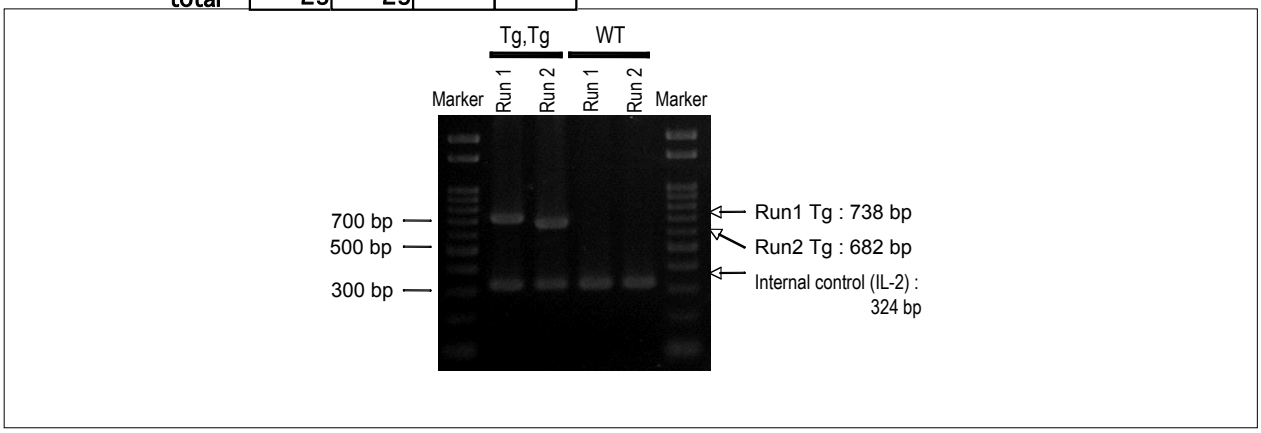
Reaction Components

Enzyme TaKaRa Ex Taq (Cat. No. RR001A)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		15.3	15.3		
PCR buffer	10 X	2.5	2.5		
dNTP	2.5 mM	2.0	2.0		
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		
primer 5	10 pmol/μl	1.0	1.0		
primer 6	10 pmol/μl	1.0	1.0		
DNA		1.0	1.0		
Taq		0.2	0.2		
	total	25	25		

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

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	738 bp	Tg (TCR α)
primer 3	primer 4	682 bp	Tg (TCR β)
primer 5	primer 6	324 bp	Internal control (IL-2)

Comment