

# PCR protocol

RBRC 01516

Strain name VChT-Cre.Slow

Primer	primer_name	primer_seq	length	
1	VChT-Cre transgene	CAGAAACCAAAGGGATTTGGAG	22	Depositor's primer
2		GAAACCTGTGAGTGTGGTGGC	21	Depositor's primer
3	Cre-coding region	ACCTGATGGACATGTTCCAGG	20	Depositor's primer
4		CGAGTTGATAGCTGGCTGG	19	Depositor's primer
5		ACCAAGAAGTGGGAAGCATC	20	Depositor's primer
6		TCCAGCAGGCCAGAGTCTC	20	Depositor's primer
7	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24	Internal control (IL-2) JAX Genotyping Protocol
8	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25	Internal control (IL-2) JAX Genotyping Protocol

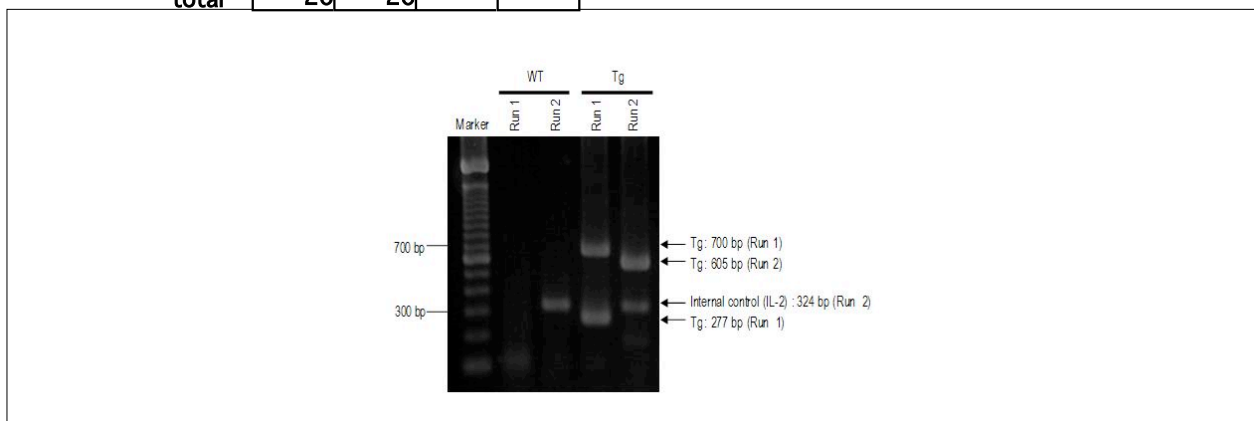
## Reaction Components

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

## Reaction Conditions

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

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		2.8	2.8		
PCR buffer	2 X GC I	10.0	10.0		
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	1.0	0		
primer 4	10 pmol/μl	1.0	0		
primer 5	10 pmol/μl	0	1.0		
primer 6	10 pmol/μl	0	1.0		
primer 7	10 pmol/μl	0	1.0		
primer 8	10 pmol/μl	0	1.0		
DNA		1.0	1.0		
Taq		0.2	0.2		
<b>total</b>		<b>20</b>	<b>20</b>		



## 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	277bp	Tg (VChT)
primer 3	700 bp	Tg (Cre)
primer 5	605 bp	Tg (ChAT)
primer 7	324 bp	Internal control (IL-2)

## Comment