PCR protocol

RBRC 02140 **Strain name** B6.Cg-Tg(DBH-tauGFP)17

Primer

	primer_name	primer_seq	length		
1	hDBH	GAAGGGACAGCATCCGCCTGTC	22		RIKEN BRC
2		TTACTTGTACAGCTCGTCC	19		Blood, 110, 107-115 (2007).
3	mLC3ex3GT	TGAGCGAGCTCATCAAGATAATCAGGT	27		Mol. Biol. Cell., 15, 1101-1111 (2004).
4	mLC3ex4AG	GTTAGCATTGAGCTGCAAGCGCCGTCT	27		Mol. Biol. Cell., 15, 1101-1111 (2004).

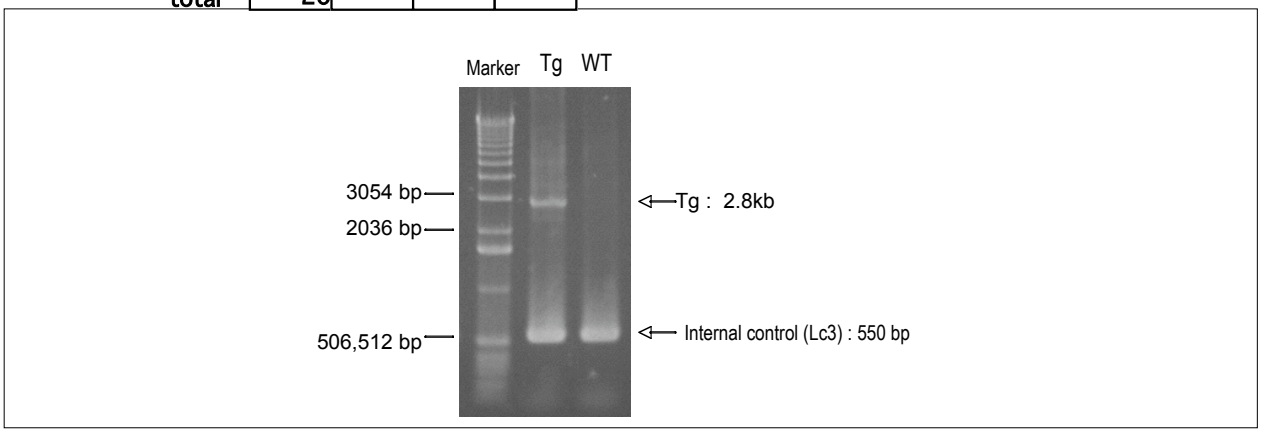
Reaction Components

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	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		0.2			
	total	20			

Reaction Conditions

step	temp (℃)	time (sec)		note
1	94	180		
2	94	30	*	
3	60	30	*	
4	72	120	*	*Repeat for 30 cycles
5	72	300		



Marker

1 kb plus DNA ladder (Invitrogen, Cat. 10787-026)

Gel

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

PCR products

primer_set		product_size	product
primer 1	primer 2	2.8 kb	Tg
primer 3	primer 4	550 bp	Internal control (LC3)

Comment