

RBRC 06355

Strain name B6.Cg-Tg(Nes-EGFP)1Yamm

Primer	primer_name	primer_seq	length	
1	NesP-S	AGTGCTCCACGTCCGCTTGC	20	Depositor's primer
2		CTTGACAGCTCGTCCATGCCGAG	24	RIKEN BRC
3	mLC3ex4AG	GTTAGCATTGAGCTGCAAGCGCCGTCT	27	Mol. Biol. Cell., 15, 1101-1111(2004).
4	mLC3ex3GT	TGAGCGAGCTCATCAAGATAATCAGGT	27	Mol. Biol. Cell., 15, 1101-1111(2004).

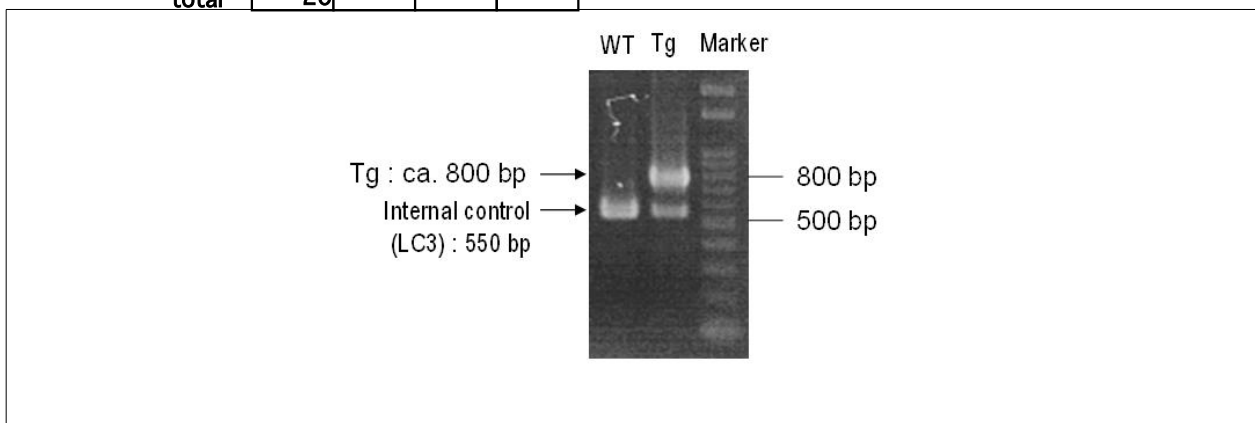
**Reaction Components**

Enzyme TOYOBO KOD FX (Code.No : KFX-101)

**Reaction Conditions**

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		0.8			
PCR buffer	2 X	10.0			
dNTP	2.0 mM	4.0			
primer 1	10 pmol/ $\mu$ l	1.0			
primer 2	10 pmol/ $\mu$ l	1.0			
primer 3	10 pmol/ $\mu$ l	1.0			
primer 4	10 pmol/ $\mu$ l	1.0			
DNA		1.0			
Taq	1 U/ $\mu$ l	0.2			
<b>total</b>		<b>20</b>			

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30*	
4	68	60*	*Repeat for 30 cycles



**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. 800 bp	Tg
primer 3 primer 4	550 bp	Internal control (LC3)

**Comment** Novagen KOD Xtreme™ Hot Start DNA Polymerase can be purchased from EMD.