

RBRC 09994

Strain name C57BL/6J-Tg(Eno2-GFP/Atg5)1Nmz

Primer	primer_name	primer_seq	length	
1	NSE-WT-F	GCAGAAAGTATCATGTCATGG	21	Depositor's primer
2	NSE-R	GAGCCGTGGGAGAGTTTTAG	20	Depositor's primer
3	GFP(LC3)1	TCCTGCTGGAGTTCGTGACCG	21	Dev. Cell, 39, 116-130 (2016).

Reaction Components

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

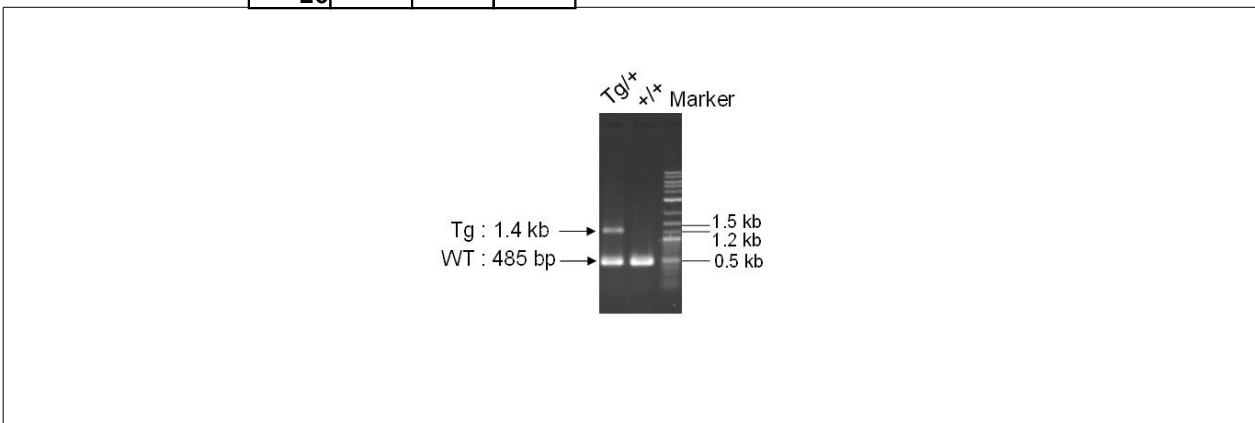
Reaction Conditions

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		1.8			
PCR buffer	2 X	10.0			
dNTP	2.0 mM	4.0			
primer 1	10 pmol/ μ l	1.0			
primer 2	10 pmol/ μ l	1.0			
primer 3	10 pmol/ μ l	1.0			
DNA		1.0			
Taq	1 U/ μ l	0.2			
total		20			

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

Zygosity determination PCR

Standard PCR



Marker

2-Log DNA Ladder (NEB, Cat. N3200)

Gel

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

Comment

PCR products	primer_set	product_size	product
primer 1	primer 2	485 bp	WT
primer 2	primer 3	1.4 kb	Tg

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