PCR protocol

RBRC 03099 **Strain name** B6.Cg-Tg(CAG-EGFP/MIR145/MIR143)B

Primer

	primer_name	primer_seq	length		
1	olMR0872	AAgTTCATCTgCACCACCg	19		JAX Genotyping Protocol
2	olMR1416	TCCTTgAAgAAgATggTgCg	20		JAX Genotyping Protocol
3	**olMR**0042	CTAGGCCACAGAATTGAAAGATCT	24		JAX Genotyping Protocol
4	**olMR**0043	GTAGGTGGAAATTCTAGCATCATCC	25		JAX Genotyping Protocol

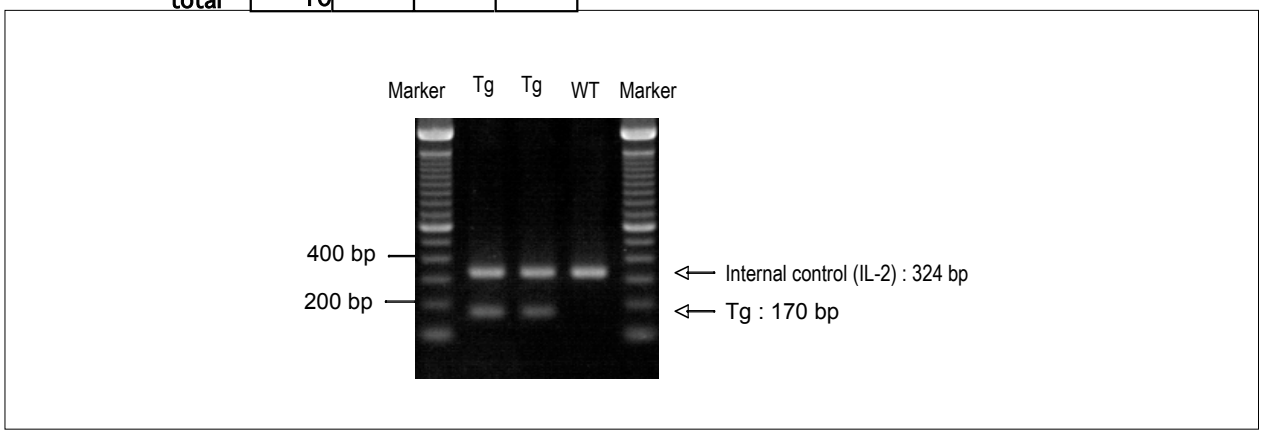
Reaction Components

Enzyme QIAGEN Multiplex PCR Kit (Cat. No. 206143)

	Conc	Run 1	Run 2	Run 3	Run 4
Master Mix	2 X	5.0			
primer 1	10 pmol/μl	1.5			
primer 2	10 pmol/μl	1.5			
primer 3	10 pmol/μl	0.5			
primer 4	10 pmol/μl	0.5			
DNA		1.0			
	total	10			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	95	900	
2	94	30*	
3	60	90*	
4	72	90*	*Repeat for 27 cycles
5	72	600	



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	170 bp	Tg
primer 3	primer 4	324 bp	Internal control (IL-2)

Comment