

# PCR protocol

RBRC 05377

Strain name C3H/HeN-Tg(CAG-CD4)17Yiw

Primer	primer_name	primer_seq	length	
1	5CAG	CCTACAGCTCCTGGGCAACGTGC	23	RIKEN BRC
2	exon 10	GCTACATGTCTTCTGAAACCGG	22	RIKEN BRC
3	oIMR0042	CTAGGCCACAGAATTGAAAGATCT	24	JAX Genotyping Protocol
4	oIMR0043	GTAGGTGGAAATTCTAGCATCATCC	25	JAX Genotyping Protocol

### Reaction Components

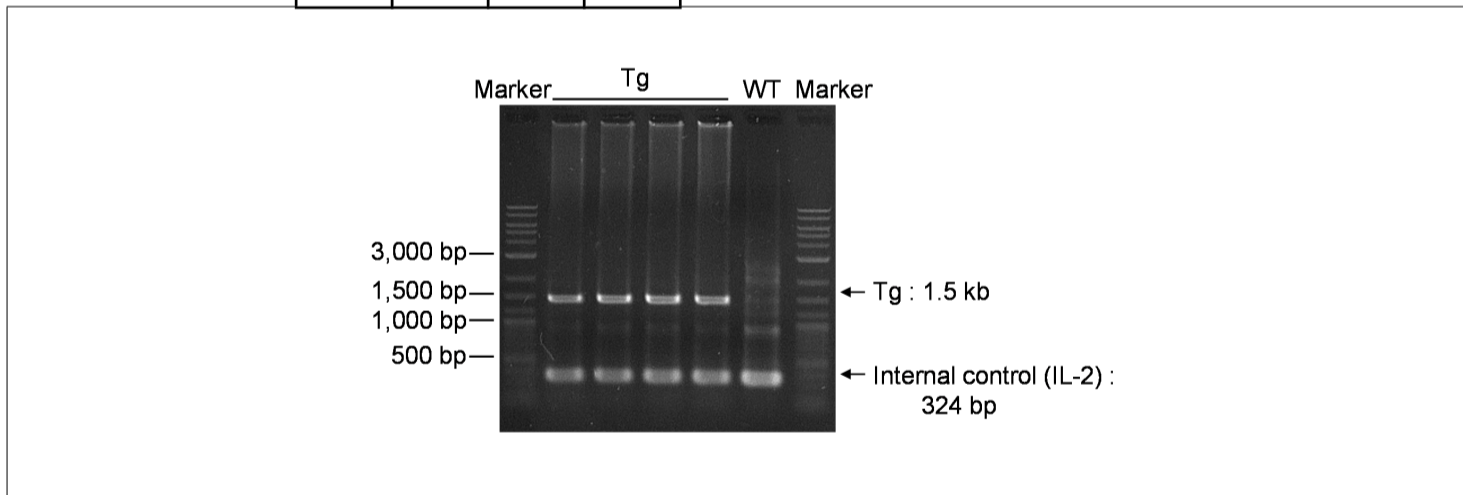
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

### Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30*	
4	68	180*	*Repeat for 30 cycles

	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>			

### Standard PCR



Marker	PCR products	primer_set	product_size	product
2-Log DNA Ladder (NEB, Cat. N3200)	primer 1	primer 2	1.5 kb	Tg
Gel	primer 3	primer 4	324 bp	Internal control (IL-2)
Comment				

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