PCR protocol

Sheet ID PS_05848
Release date : 6/9/2015
Print date : 6/9/2015

RBRC 06317 **Strain name** C.Cg-Cables1<Tg(CAG-rtTA,CAG-tTS)138Utr>

Primer	primer_name	primer_seq	length		
1	TAS2	ATACCATGGGGACAGATGGA	20		Depositor's primer
2	TAS3	ACAGCGAAGACGCCACTTAT	20		Depositor's primer
3	oIMR8746	CGCTGTGGGGCATTTTACTTTAG	23		JAX Genotyping Protocol
4	tTA5'	AAGTGATTAACAGCGCATTAG	21		

Reaction Components

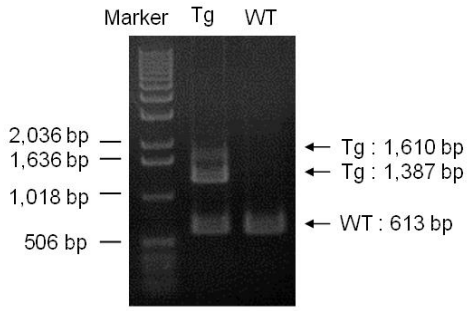
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

	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/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	1 U/μl	0.2			
	total	20			

Reaction Conditions

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

Standard PCR



Marker

1 kb DNA ladder (Invitrogen, Cat. 15615-024)

Gel

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

PCR products

primer_set		product_size	product
primer 1	primer 2	613 bp	WT
primer 1	primer 3	1,387 bp	Tg
primer 1	primer 4	1,610 bp	Tg

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

PCR protocol

Sheet ID PS_05845
Release date : 5/21/2014
Print date : 6/9/2015

RBRC 06317 Strain name C.Cg-Cables1<Tg(CAG-rtTA,CAG-tTS)138Utr>

Primer	primer_name	primer_seq	length		
1	TAS1	ATATGAAACAGCCCCCTGCT	20		Depositor's primer
2	TAS2	ATACCATGGGGACAGATGGA	20		Depositor's primer
3	TAS3	ACAGCGAAGACGCCACTTAT	20		Depositor's primer

Reaction Components

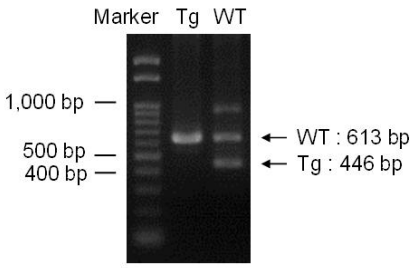
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

	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/μl	1.0			
primer 2	10 pmol/μl	1.0			
primer 3	10 pmol/μl	1.0			
DNA		1.0			
Taq	1 U/μl	0.2			
	total	20			

Reaction Conditions

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

Standard PCR



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	446 bp	Tg
primer 2	primer 3	613 bp	WT

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