PCR protocol

Sheet ID PS_06493
Release date : 8/14/2015
Print date : 8/14/2015

RBRC 00858 Strain name B6;129S6-Bcl2l12/Irf3<tm1Ttg>/TtgRbrc

Primer	primer_name	primer_seq	length		
1		GTGAGGCGGTTGTGAGAGCAG	21		RIKEN BRC
2		GTCAGCCATCTGTGCGTCCTGC	22		RIKEN BRC
3		AAGAACTCGTCAAGAAGGCGATAGAAGGCG	30		

Reaction Components

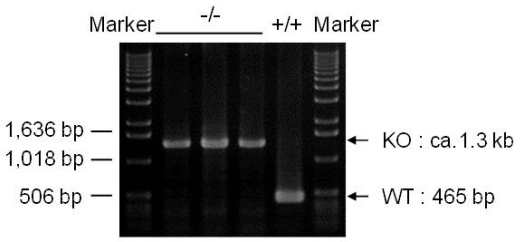
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

	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	120	* *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	465 bp	WT
primer 2	primer 3	ca 1.3 kb	KO

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

PCR protocol

Sheet ID PS_05140
Release date : 6/09/2015
Print date : 8/14/2015

RBRC 00858 Strain name IRF-3 Knockout mice, C57BL/6J

Primer	primer_name	primer_seq	length		
1	IRF-3 common	GAACCTCGGAGTTATCCCGAAGG	23		Depositor's primer
2	IRF-3 d.s	GTTTGAGTTATCCCTGCACTTGGG	24		Depositor's primer
3		TCGTGCTTTACGCTATCGCCGCTCCCGATT	30		Depositor's primer

Reaction Components

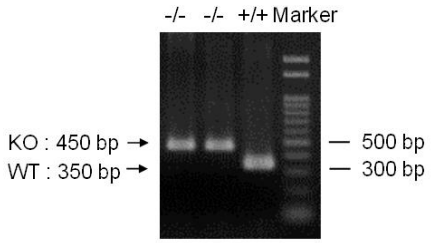
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		1.8			
PCR buffer	2 X	10.0			
dNTP	2 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 3	450 bp	KO
primer 1	primer 2	350 bp	WT

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

PCR protocol

Sheet ID PS_00176
Release date : 6/1/2006
Print date : 8/14/2015

RBRC 00858 Strain name IRF-3 Knockout mice

Primer	primer_name	primer_seq	length		
1	IRF-3 common	GAACCTCGGAGTTATCCCGAAGG	23		Depositor's primer
2	IRF-3 d.s	GTTTGAGTTATCCCTGCACTTGGG	24		Depositor's primer
3	IRF-3 mut	TCGTGCTTTACGCTATCGCCGCTCCCGATT	30		Depositor's primer

Reaction Components

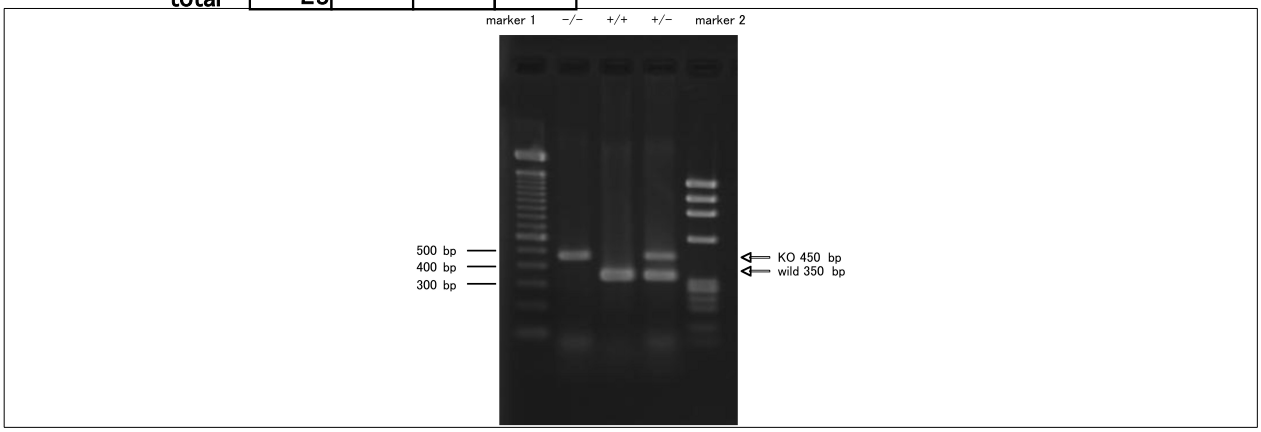
Enzyme: Roche Taq (Cat. No. BM1418432)

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		16.3			
PCR buffer	10 X	2.5			
dNTP	2.5 mM	2.0			
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		0.2			
	total	25			

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	180	
2	94	30	*
3	60	30	*
4	72	60	* *Repeat for 32 cycles
5	72	300	

Standard PCR



Marker

marker 1:100 bp ladder (Invitrogen, Cat. 15628-050), marker2: ϕX174 Hae III

Gel

2% agarose in 1 X TAE (100 V, 30 min)

PCR products

primer_set		product_size	product
primer 1	primer 3	450 bp	KO
primer 1	primer 2	350 bp	WT

Comment