

RBRC 06297

Strain name CAJcl.Cg-Aicda<tm1Hon>

Primer	primer_name	primer_seq	length
1	238	CTGCCAAACCTGATGTCTTGAGTTTGAT	28
2	AID-creWAS	TAGCAGAGGTAGGTCTCATGCCGTCCCT	28
3	Neo10	ATCAGAGCAGCCGATTGTCTGTTG	24

Reaction Components

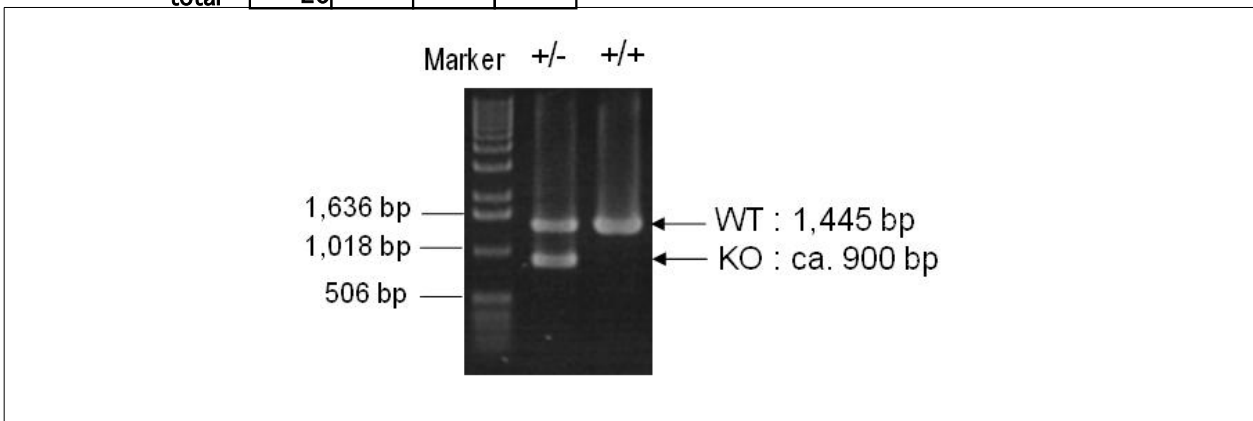
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

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	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		product_size	product
primer 1	primer 2		
primer 1	primer 2	1,445 bp	WT
primer 1	primer 3	ca. 900 bp	KO

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

RBRC 06297

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Primer	primer_name	primer_seq	length	
1	227	CAACGTGGCGTCCAAACAGGCACTTCCG	28	
2	AID-R1	CCAGGCTTTGAAAGTTCTTTCACG	24	
3	5Neo	TGGATTGCACGCAGGTTCTC	20	Proc. Natl. Acad. Sci. USA, 94, 13069-13074 (1997).

Reaction Components

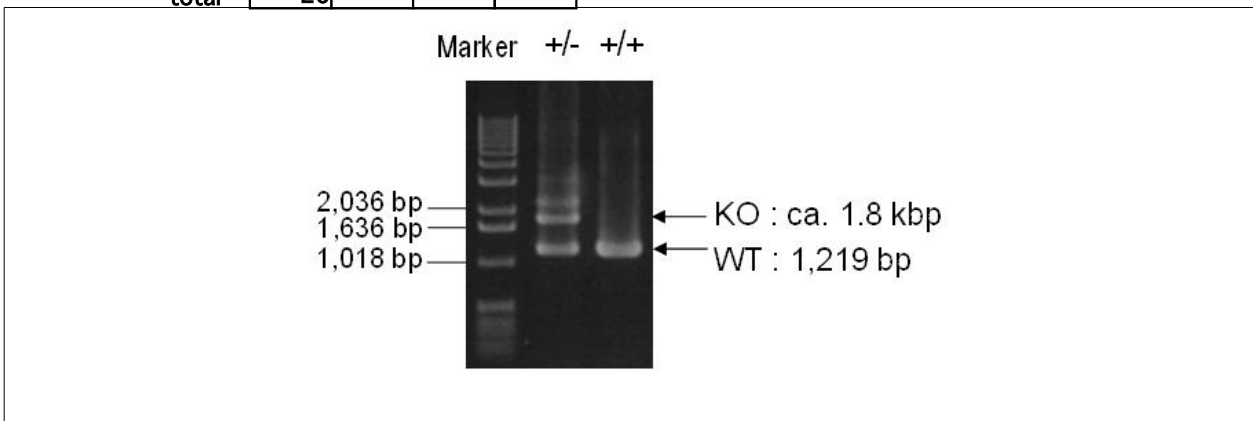
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

Reaction Conditions

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DNA		1.0			
Taq	1 U/ μ l	0.2			
total		20			

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	1,219 bp	WT
primer 1 primer 3	ca. 1.8 kbp	KO

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