

RBRC 03666

Strain name Arx-6 KO/GAD67 GFPdNeo KI

Primer	primer_name	primer_seq	length	
1	mArx fff	CGCCCAAGGAAGAGCTGTTGCTGC	24	Depositor's primer
2	mArx eer	TATTCCACCCTCCTGGACCCTTTC	24	Depositor's primer
3	pMC1neo stop f	GCCTTCTTGACGAGTTCTTC	20	Depositor's primer
4		CCTTCTGTCTCCGAGACTTCGGTG	24	RIKEN BRC
5		GCGCAGCCTCCCGCAGTGCCAGC	23	RIKEN BRC
6	oIMR1416	GCAGAAGAACGGCATCAAGGTG	22	RIKEN BRC

Reaction Components

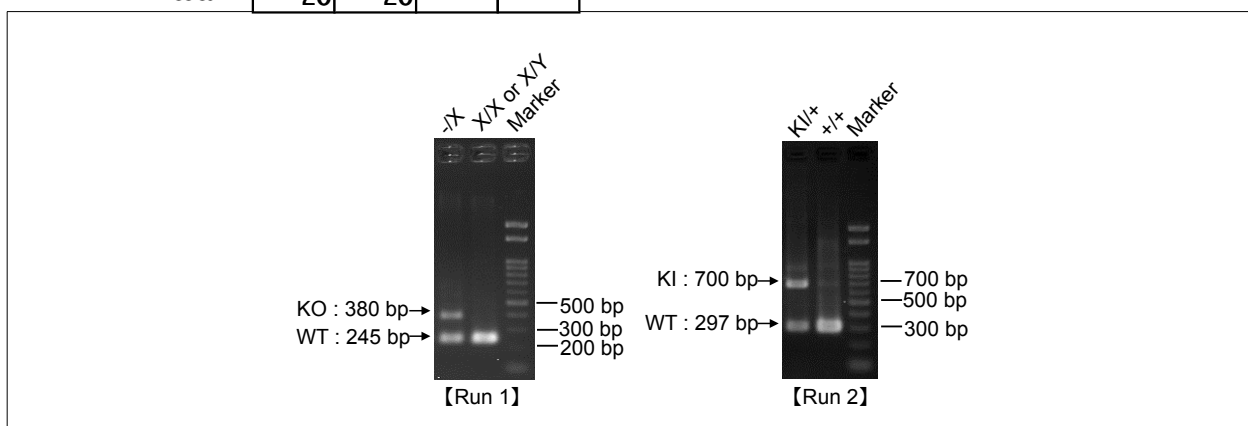
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

Reaction Conditions

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

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		1.8	1.8		
PCR buffer	2 X	10.0	10.0		
dNTP	2.0 mM	4.0	4.0		
primer 1	10 pmol/μl	1.0	0		
primer 2	10 pmol/μl	1.0	0		
primer 3	10 pmol/μl	1.0	0		
primer 4	10 pmol/μl	0	1.0		
primer 5	10 pmol/μl	0	1.0		
primer 6	10 pmol/μl	0	1.0		
DNA		1.0	1.0		
Taq	1 U/μl	0.2	0.2		
total		20	20		

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	245 bp	WT
primer 2	primer 3	380 bp	KO
primer 4	primer 5	297 bp	WT
primer 5	primer 6	700 bp	KI

Comment

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