PCR protocol

Sheet ID PS_08577
Release date : 12/25/2023
Print date : 12/25/2023

RBRC 06294

Strain name B6;129-Aqp7<tm1Tfun>

Primer	primer_name	primer_seq	length		Reference
1	promoter	CACCCGCTTCCATCTTGTCAGC	22		RIKEN BRC
2	promoter	CCTAAGCAAGGAACACGGTAGAG	23		RIKEN BRC
3	neo_F	GCAGCGCATCGCCTTCTATCGC	22		RIKEN BRC

Reaction Components

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

Reagent	Conc	Reaction 1	2	3	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			

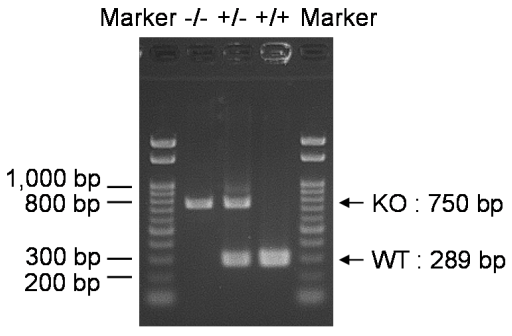
(μL)

Reaction Conditions

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

Marker Gene Ladder 100 (Wako, Cat. 316-06951)

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



primer_set		product_size	product (allele type)
primer 1	primer 2	289 bp	WT
primer 1	primer 3	750 bp	KO

Comment Standard PCR

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

PCR protocol

Sheet ID PS_05864
Release date : 5/22/2014
Print date : 12/25/2023

RBRC 06294

Strain name B6;129-Aqp7<tm1Tfun>

Primer	primer_name	primer_seq	length		Reference
1	ex3_Fw	CTTGGTCTGCTGCTTCAGGTC	21		RIKEN BRC
2	Rv	CAGAGTCCCTCTCACGTCAC	20		RIKEN BRC
3	Neo	GCAATCCATCTTGTTCAATGGCC	23		Depositor's primer

Reaction Components

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

Reagent	Conc	Reaction 1	2	3	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			

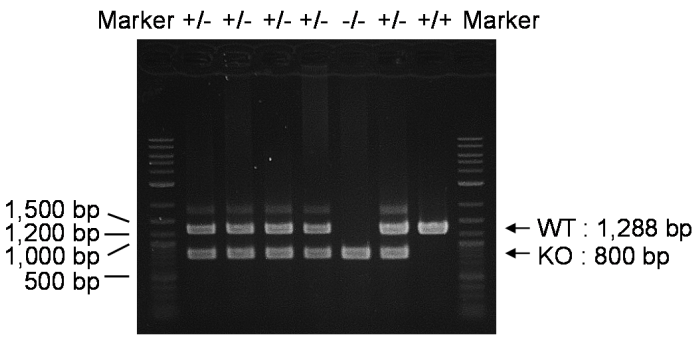
(μL)

Reaction Conditions

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

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

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



primer_set		product_size	product (allele type)
primer 1	primer 2	1,288 bp	WT
primer 2	primer 3	ca 800 bp	KO

Comment Standard PCR

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