PCR protocol

Sheet ID PS_06053
Release date : 5/6/2022
Print date : 5/6/2022

RBRC 06260

Strain name C57BL/6N-Atp6v0a4<tm1(KOMP)Vlcg>/AB3

Primer	primer_name	primer_seq	length		Reference
1	TDF	GCTTTCCCTAGCAGATGTGAC	21		http://www.velocigene.com/
2	SD	CACAACCAGTTCAGCTACTG	20		http://www.velocigene.com/
3	Neo_F	GGTGGAGAGGCTATTCGGCTATG	23		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			

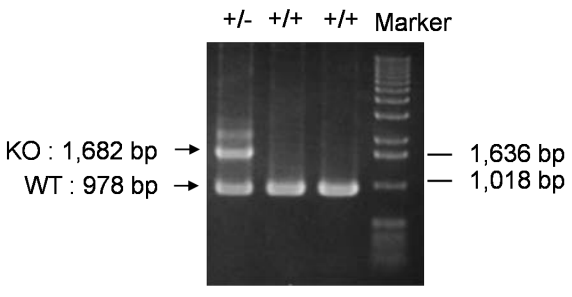
(μL)

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30*	
4	68	180*	*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	978 bp	WT
primer 2	primer 3	1,682 bp	KO

Comment Standard PCR

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

PCR protocol

Sheet ID PS_06052
Release date : 12/11/2014
Print date : 5/6/2022

RBRC 06260

Strain name C57BL/6N-Atp6v0a4<tm1(KOMP)Vlcg>/AB3

Primer	primer_name	primer_seq	length		Reference
1	SU	TTTGGGTTTGATGATGTTGG	20		http://www.velocigene.com/
2	TUR	GTGCTTGTTAGTCTTGCTTGTTAGG	25		http://www.velocigene.com/
3	lacZ-R	CGCATCGTAACCGTGCATCTGCC	23		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			

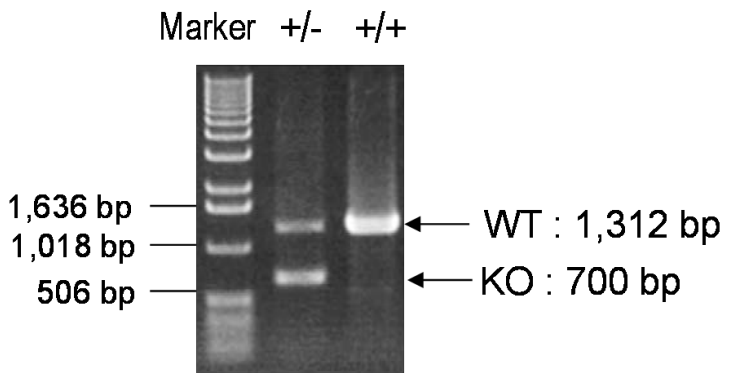
(μL)

Reaction Conditions

step	temp (℃)	time (sec)	note
1	94	120	
2	98	10*	
3	60	30*	
4	68	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	1,312 bp	WT
primer 1	primer 3	700 bp	KO

Comment Standard PCR

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