

RBRC 11536

Strain name B6-Aldh3b2<em1Akih>

Primer	primer_name	primer_seq	length	Reference
1	wild	CAGCTGCGGAGCCTGGGTCGC	21	RIKEN BRC
2	Aldh3b2-2	CAGTCTGTGCATAGAGCAAGGG	22	Mol Cell Biol., 41(10):e0035221 (2021).
3	intron 1	CTACCAATGAACACTGTCCCATCC	24	RIKEN BRC
4	mut	GCAGCTCTTTGTTTTCTGCAACC	24	RIKEN BRC

Reaction Components

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

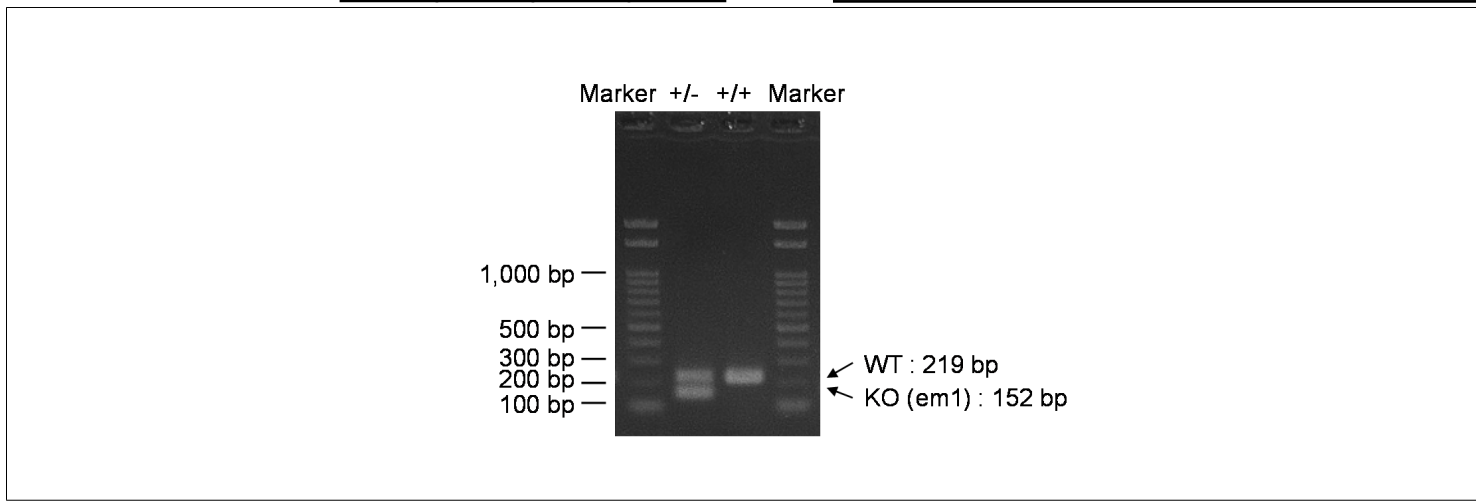
Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	65	30*	
4	68	60*	*Repeat for 30 cycles

Reagent	Conc	Reaction 1	2	3	4
H2O		0.8			
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			
primer 4	10 pmol/μl	1.0			
DNA		1.0			
Taq	1 U/μl	0.2			
total		20			

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	219 bp	WT
primer 3	primer 4	152 bp	KO (em1)

+/+ (wild-type): 219 bp
 +/- (heterozygous): 219 bp and 152 bp

Comment Allele specific PCR (ASP)

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