

RBRC 10212

Strain name B6;Cg-Paxbp1 <tm1.1Hko>

Primer	primer_name	primer_seq	length	Reference
1	Gcfc1-U22200	AGAGAGATTTCTGTTTGAGCC	22	Depositor's primer
2	Gcfc1-L22347	ACCAACATCAACTTCTTACGC	21	Depositor's primer

Reaction Components

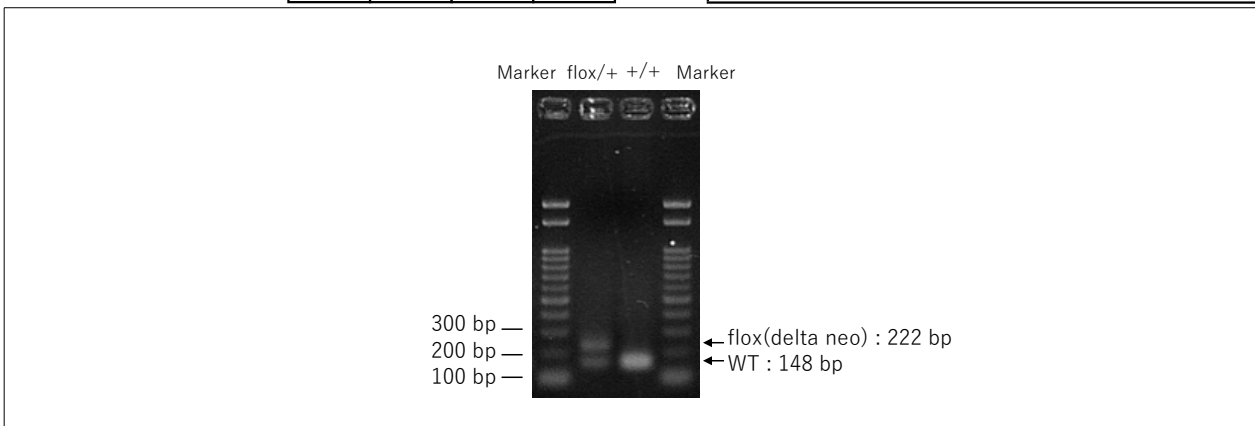
Enzyme TOYOBO KOD FX (Code.No : KFX-101)

Reaction Conditions

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

Reagent	Conc	Reaction 1	2	3	4
H2O		2.8			
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			
DNA	10 pmol/ μ l	1.0			
Taq	1 U/ μ l	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	148 bp	WT
primer 1	222 bp	flox delta neo

Comment [Standard PCR](#)

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

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Primer	primer_name	primer_seq	length	Reference
1	Gcfc1-U22200	AGAGAGATTTCTGTTTGAGCC	22	Depositor's primer
2	Gcfc1-U19925	GGAACAACACTGGAGTCGGAG	21	Depositor's primer
3	Gcfc1-L22347	ACCAACATCAACTTCTTACGC	21	Depositor's primer

Reaction Components

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

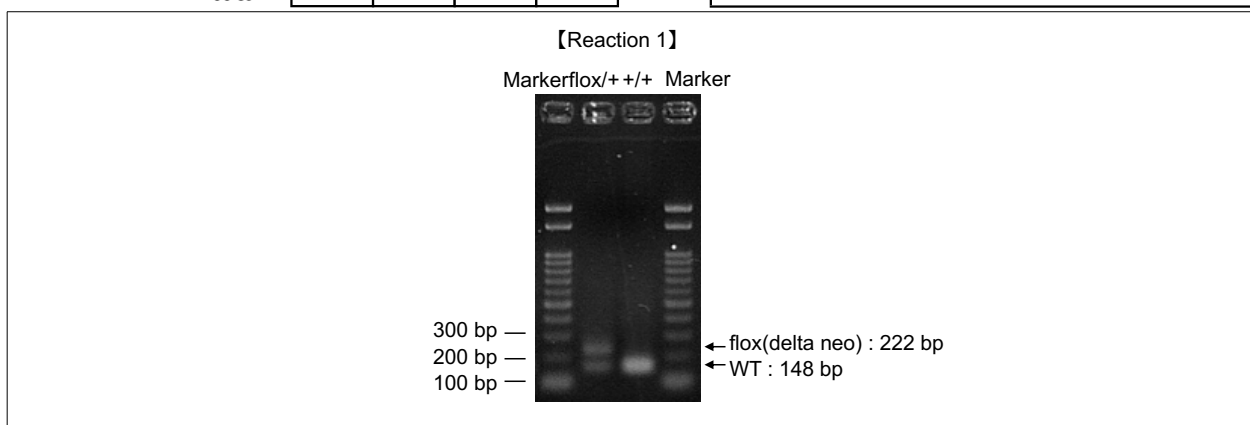
Reagent	Conc	Reaction 1	2	3	4	(μ L)
H2O		2.8	2.8			
PCR buffer	2 X	10.0	10.0			
dNTP	2.0 mM	4.0	4.0			
primer 1	10 pmol/ μ l	1.0	0			
primer 2	10 pmol/ μ l	0	1.0			
primer 3	10 pmol/ μ l	1.0	1.0			
DNA		1.0	1.0			
Taq	1 U/ μ l	0.2	0.2			
total		20	20			

Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	[Reaction 1]
2	98	10*	
3	60	30*	
4	68	60*	*Repeat for 30 cycles
1	94	120	[Reaction 2]
2	98	10*	
3	60	30*	
4	68	180*	*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 3	148 bp	WT
primer 1	primer 3	222 bp	flox delta neo
primer 2	primer 3	2,423 bp	WT
primer 2	primer 3	2,531 bp	flox delta neo
primer 2	primer 3	233 bp	null (After cre mediated excision)

[Reaction 1]
 +/- : 148 bp
 flox delta neo/+ : 148 bp, 220 bp

[Reaction 2]
 +/- : 2423 bp
 flox delta neo/+ : 2423 bp, 2531 bp
 +/- : 2423 bp, 233 bp

Comment Standard PCR

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