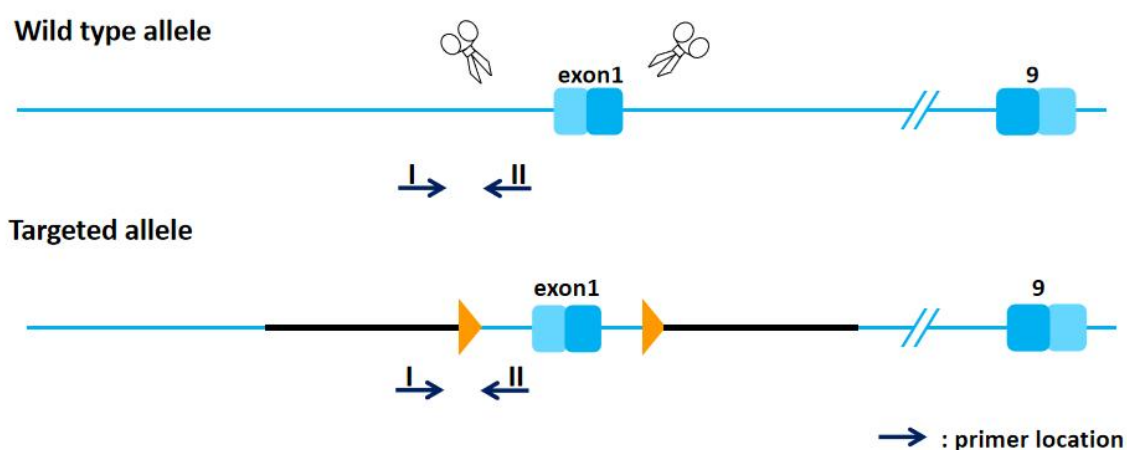


Cibar2-Flox Genotyping Protocol



Common Name	Cibar2-Flox	Cat. NO.	NM-CK0-234077
Strain of Origin	C57BL/6J	Version	V1

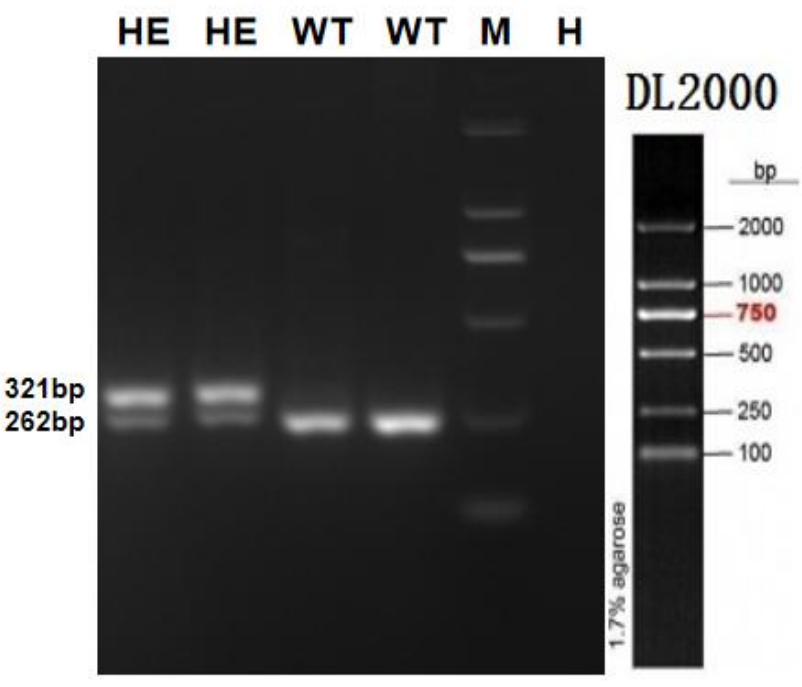
Genotyping strategy



Primers

Primer	Sequence (5' → 3')	Primer type
P1	ACAGATACCACAGGGACCCA	Forward
P2	TGCACATCTTGCTGAGCCAT	Reverse

Expected results

Results	 HE HE WT WT M H DL2000 bp 2000 1000 750 500 250 100 1.7% agarose 321bp 262bp
Genotype	Wild type: P1P2 =262 bp; Heterozygote: P1P2 =321 bp and 262 bp; Homozygote: P1P2 =321 bp

Reaction &Cycling

PCR Reaction System	Reaction Component	Volume (μl)
	ddH2O	8.0
	2×Taq Plus Master Mix	10.0
	P1 (10 pmol/μl)	0.5
	P2 (10 pmol/μl)	0.5
	Genomic DNA	1.0
	Total	20

	2×Taq Plus Master Mix from Vazyme (Code Number: P222-1)			
Cycling Reaction	Step	Temp	Time	Note
	1	95° C	5 min	
	2	95° C	20 sec	
	3	60° C	20 sec	
	4	72° C	20 sec	35 repeats to 2
	5	72° C	5 min	
	6	12° C	Hold	