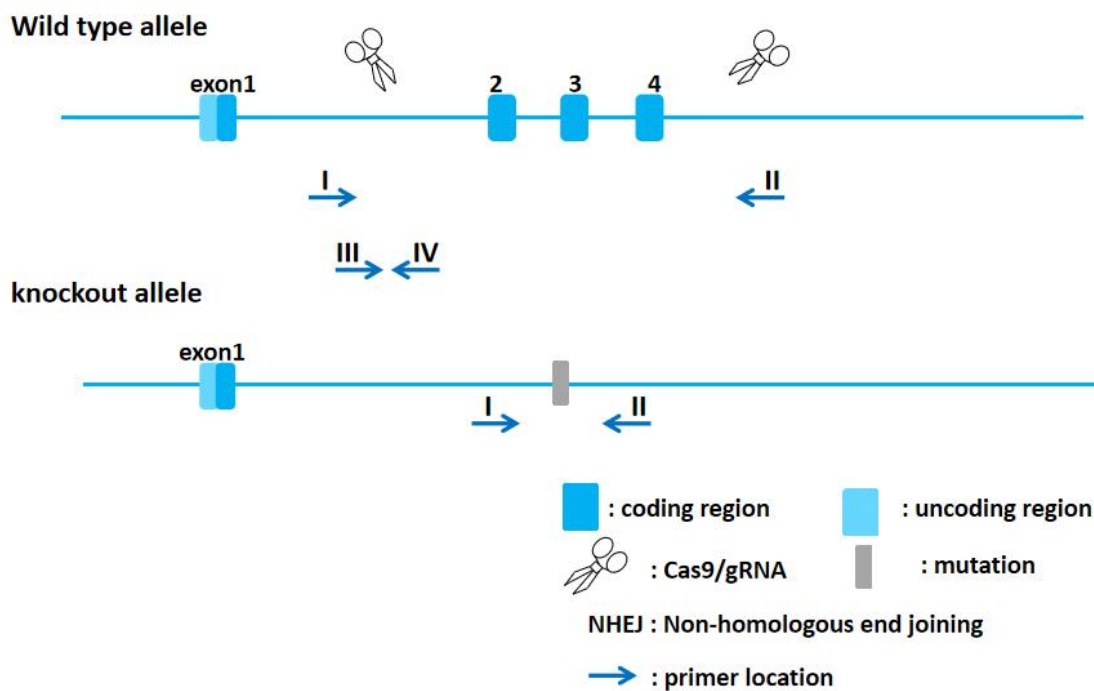


Cd300ld-KO Genotyping Protocol



Common Name	Cd300ld-KO	Cat. NO.	NM-KO-242280
Strain of Origin	C57BL/6J	Version	V1

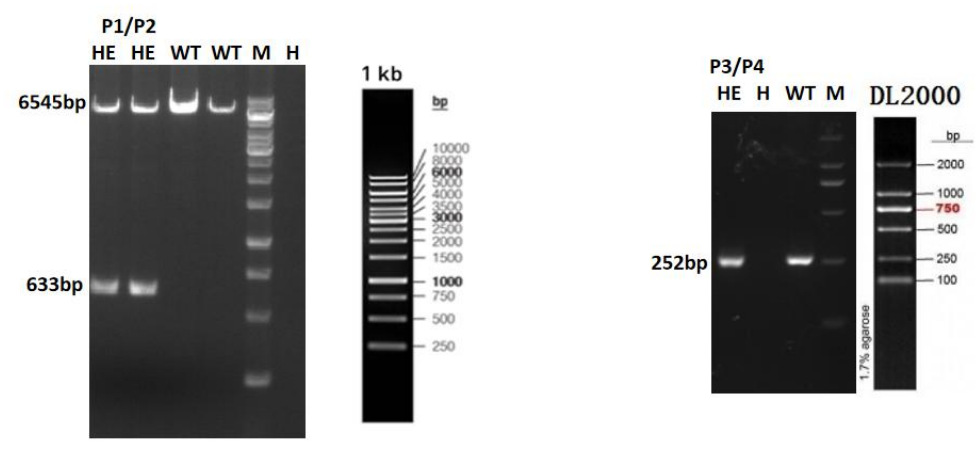
Genotyping strategy



Primers

Primer	Sequence (5'→3')	Primer type
P1	GTGTAAGCCCTCACCAACA	Forward
P2	ACAGAAGGGCCATGCTAGAC	Reverse
P3	CACGGATCTGAAGGACAGCA	Forward
P4	TGACCAATGTCTGGAATAGGGG	Reverse

Expected results

Results	
Genotype	Knockout type: -5912 bp Wild type: P1P2 =6545 bp; P3P4 =252 bp Heterozygote: P1P2 =6545 bp and 633 bp; P3P4 =252 bp Homozygote: P1P2 =633 bp

Note: In both wild-type and heterozygous mice, whether the P1 and P2 primers can amplify larger bands does not affect the interpretation of the results, because the purpose of designing this pair of primers is to amplify KO band.

Reaction &Cycling

PCR Reaction System	Reaction Component			Volume (μl)
	ddH ₂ O			8.0
	2 × Rapid Taq Master Mix			10.0
	P1(10 pmol/μl) or P3(10 pmol/μl)			0.5
	P2(10 pmol/μl) or P4(10 pmol/μl)			0.5
	Genomic DNA			1.0
	Total			20
	2 × Rapid Taq Master Mix from Vazyme(Code Number: P222-01)			
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	