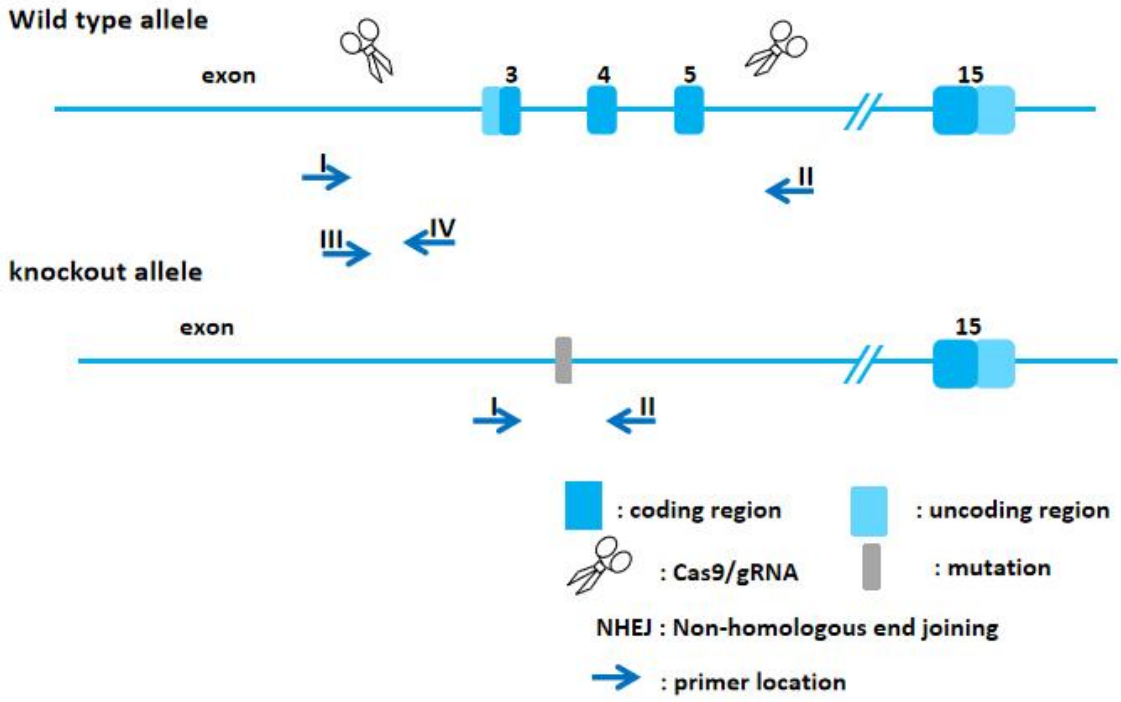
SHANGHAI MODEL ORGANISMS

Editing Genes, Decoding the Mystery of Life

Cd22-KO(2) Genotyping Protocol

Common Name	Cd22-KO(2)	Cat. NO.	NM-KO-232672
Strain of Origin	C57BL/6J	Version	V1

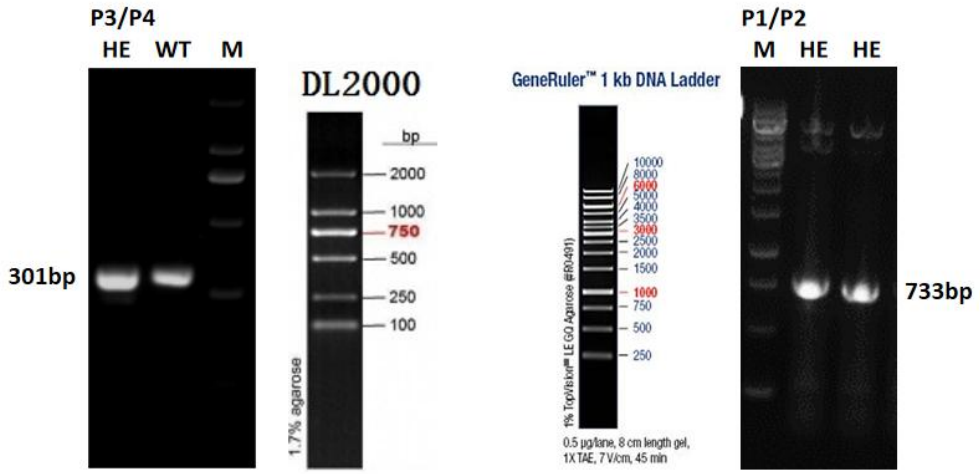
Genotyping strategy



Primers

Primer	Sequence (5' →3')	Primer type
P1	GGGAGGTGAGGATGCTGTGC	Forward
P2	ATGGATTGATTGATGCTGGAGGT	Reverse
P3	GTTGCCCTTAAATTGATCCGTCAG	Forward
P4	GGGGCAGGCATCCCTTTTCTA	Reverse

Expected results

Results	
Genotype	Knockout type: -2981 bp Wild type: P3P4 =301 bp Heterozygote: P1P2 =733 bp; P3P4 =301 bp Homozygote: P1P2 =733 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 Component	Volume (μl)
Reaction	ddH20	8.0
System	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	