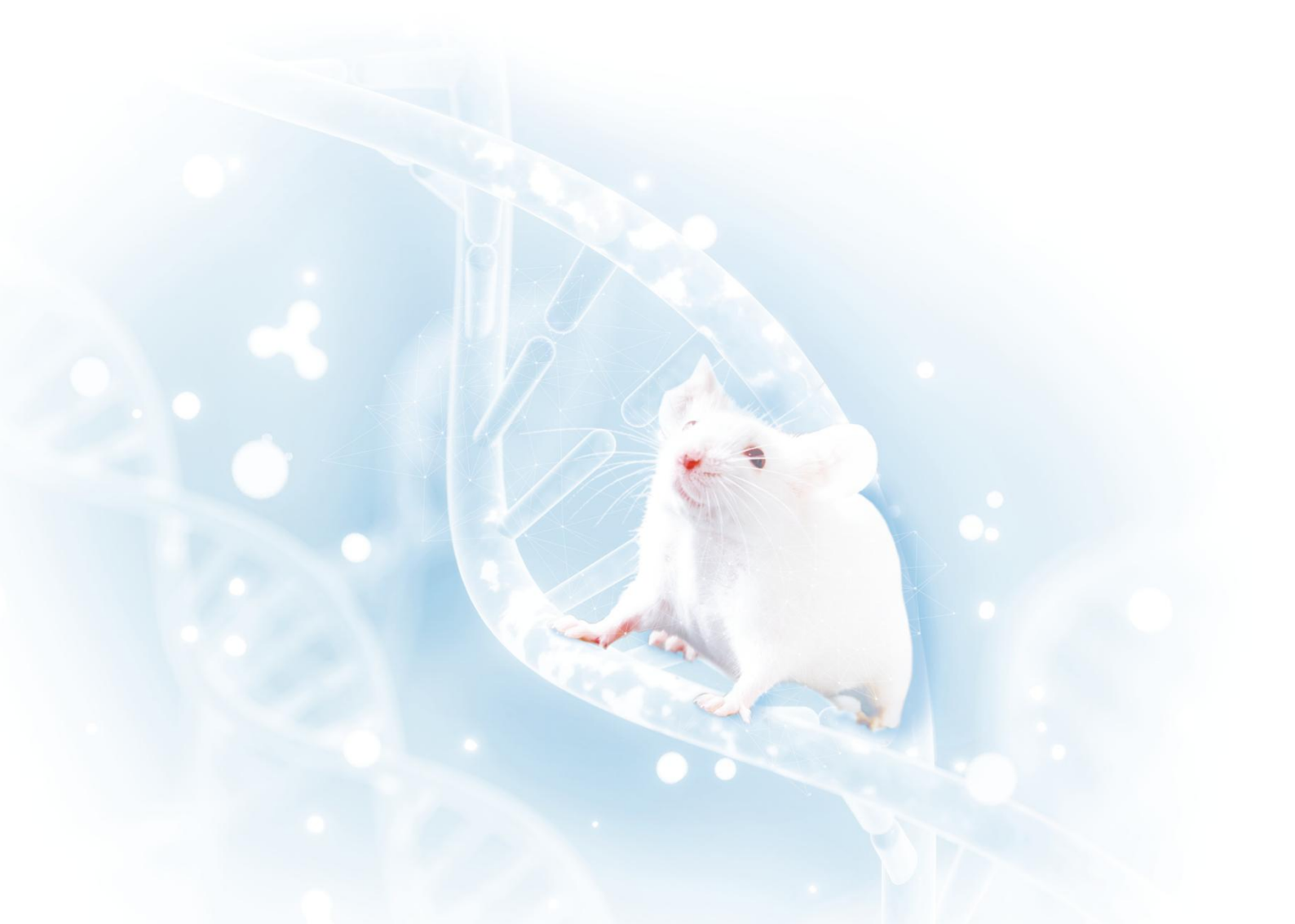
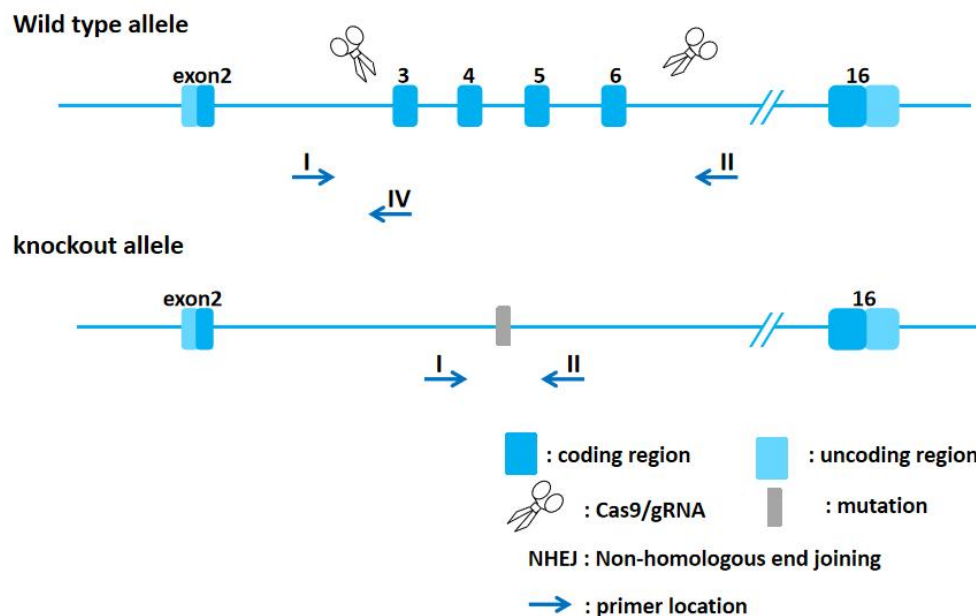


Abat-KO Genotyping Protocol



Common Name	Abat-K0	Cat. NO.	NM-K0-231646
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy



Primers

Primer	Sequence (5' → 3')	Primer type
P1	TACAACCTCATTGGCCCTGG	Forward
P2	TCTTAGTTTATGGGTGTGCCTGAA	Reverse
P4	AATGTCAACTTTGGCAGCGG	Reverse

Expected results

Results	
Genotype	Knockout type: -10982 bp Wild type: P1P4 =257 bp Heterozygote: P1P2 =500 bp; P1P4=257 bp Homozygote: P1P2 =500 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)
	ddH2O			7.5
	2×Taq Plus Master Mix			10.0
	P1 (10 pmol/μl)			0.5
	P2 (10 pmol/μl)			0.5
	P4 (10 pmol/μl)			0.5
	Genomic DNA			1.0
	Total			20
Cycling Reaction	2×Taq Plus Master Mix from Vazyme (Code Number: P222-1)			
	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	