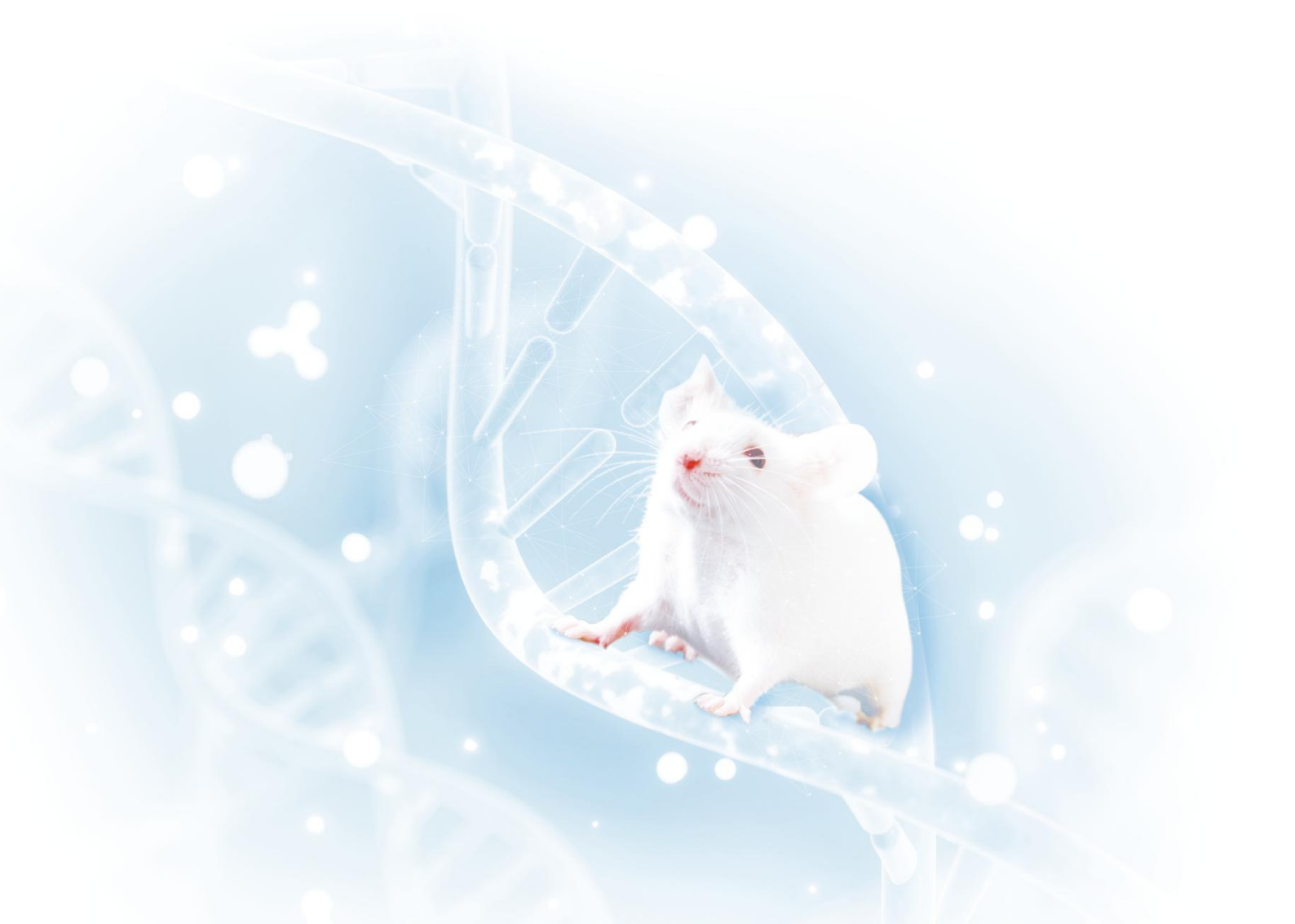
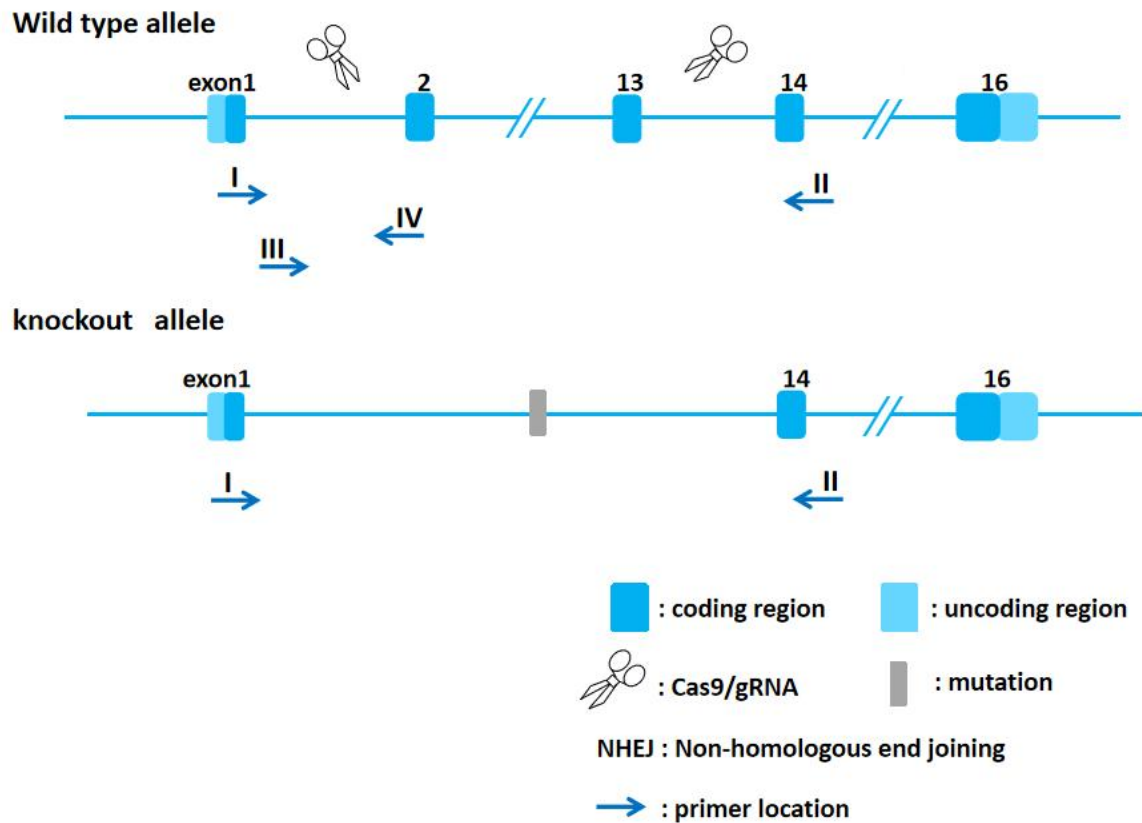


Aaas-KO Genotyping Protocol



Common Name	Aaas-KO	Cat. NO.	NM-KO-240292
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy



Primers

Primer	Sequence (5'→3')	Primer type
P1	TGACCGGCAACAGCTATGAG	Forward
P2	TCAGCTTGCACTTACCAGGG	Reverse
P3	AGTGATTGGGAACGGCATGA	Forward
P4	GGGGCTTTTATGGGATCCTT	Reverse

Expected results

Results	
Genotype	<p>Knockout type: -11217bp</p> <p>Wild type: P1P2 =11741 bp; P3P4 =391 bp</p> <p>Heterozygote: P1P2 =11741 bp and 524 bp; P3P4 =391 bp</p> <p>Homozygote: P1P2 =524 bp</p>

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	35 repeats to 2
	2	95°C	20 sec	
	3	60°C	20 sec	
	4	72°C	20 sec	
	5	72°C	5 min	
	6	12°C	Hold	