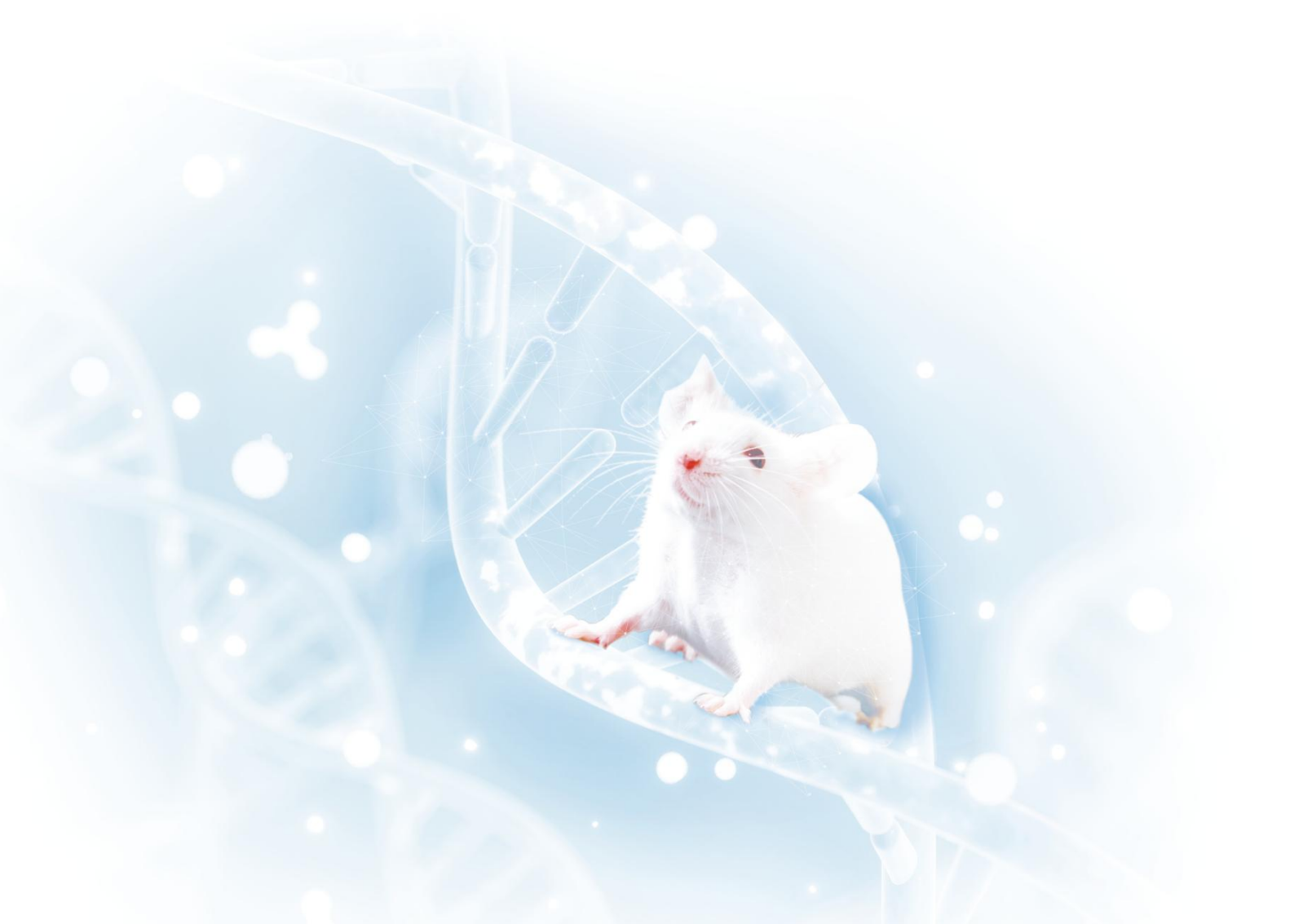
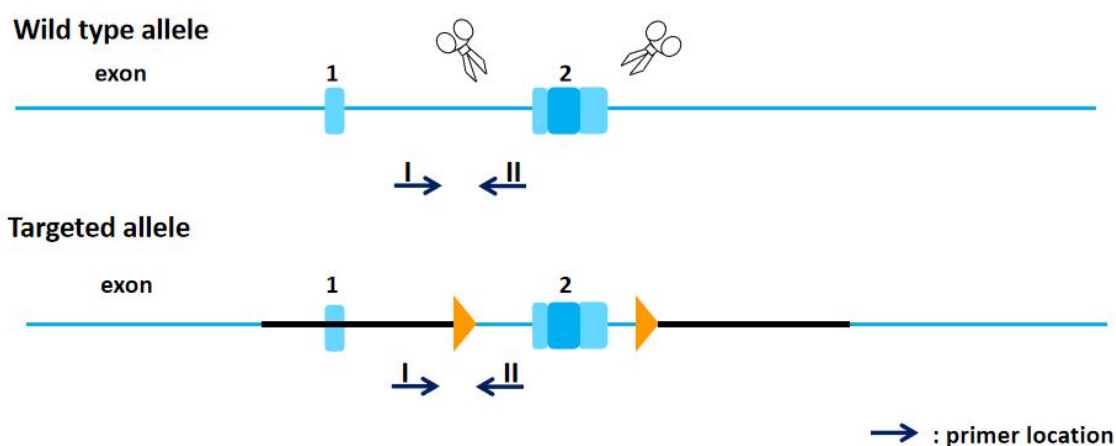


Tob1-Flox Genotyping Protocol



Common Name	Tob1-Flox	Cat. NO.	NM-CK0-234772
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy



Primers

Primer	Sequence (5' → 3')	Primer type
P1	GCAAGAGTGTGAGAGCCCTT	Forward
P2	TACTGCACACTCATCCACCC	Reverse

Expected results

Results	HE HE WT M H 349bp 290bp DL2000 bp 2000 1000 750 500 250 100 1.7% agarose
Genotype	Wild type: P1P2 =290 bp; Heterozygote: P1P2 =349 bp and 290 bp; Homozygote: P1P2 =349 bp

Reaction &Cycling

PCR Reaction System	Reaction Component	Volume (μl)
	ddH2O	8.0
	2 × Rapid Taq Master Mix	10.0
	P1 (10 pmol/μl)	0.5
	P2 (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	