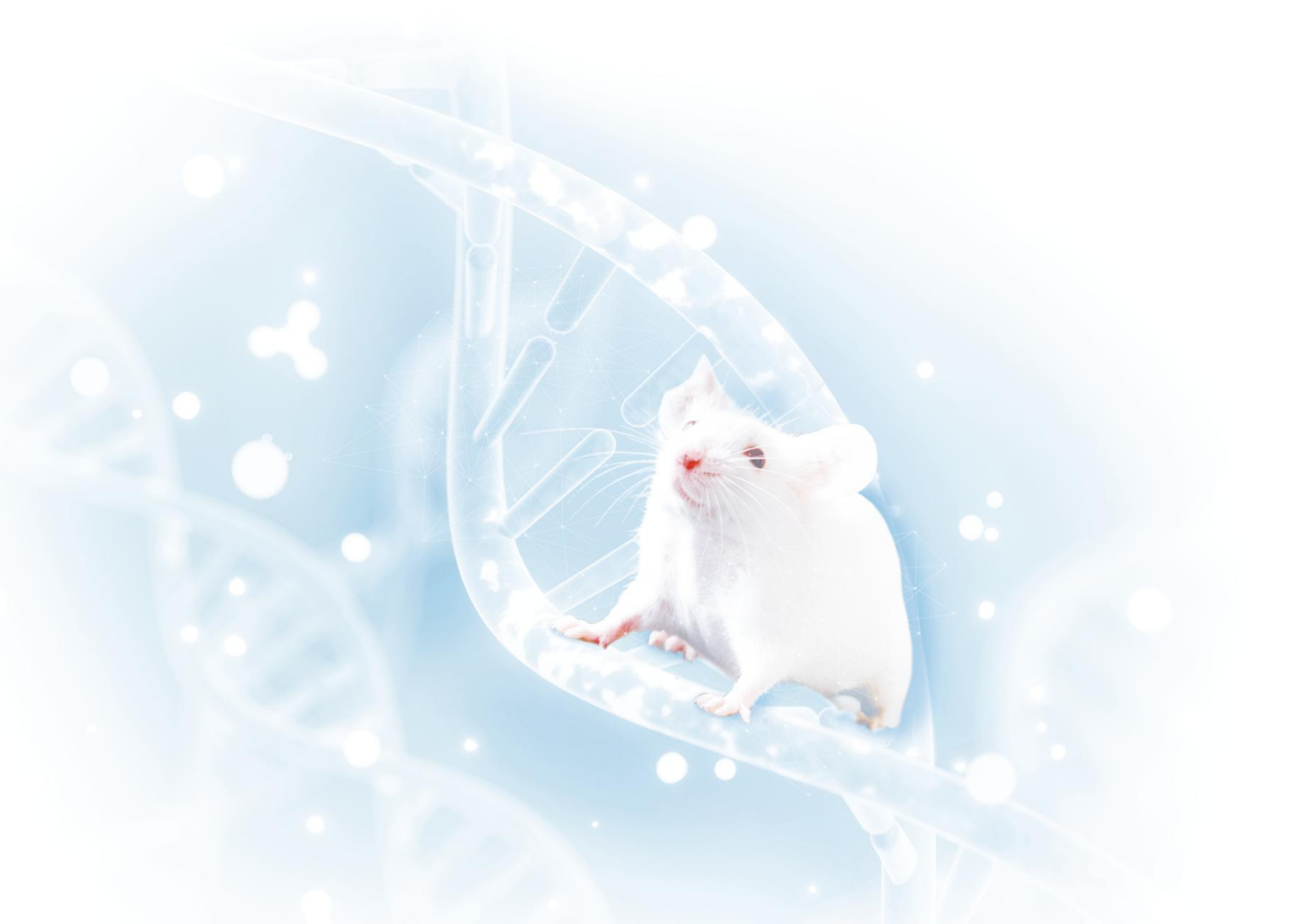


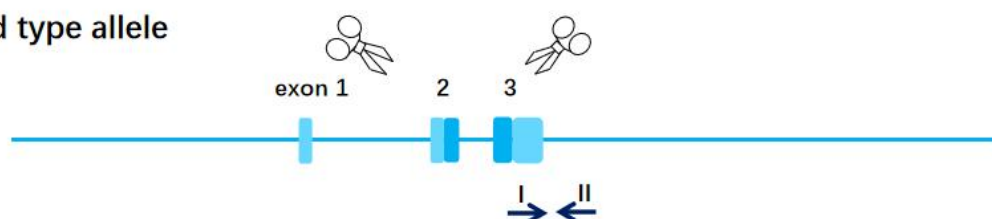
Trh-Flox Genotyping Protocol



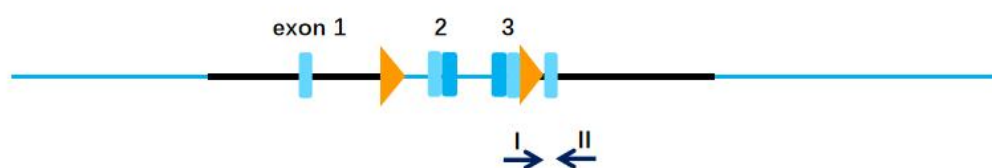
Common Name	Trh-Flox	Cat. NO.	NM-CK0-240054
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy

Wild type allele



Targeted allele

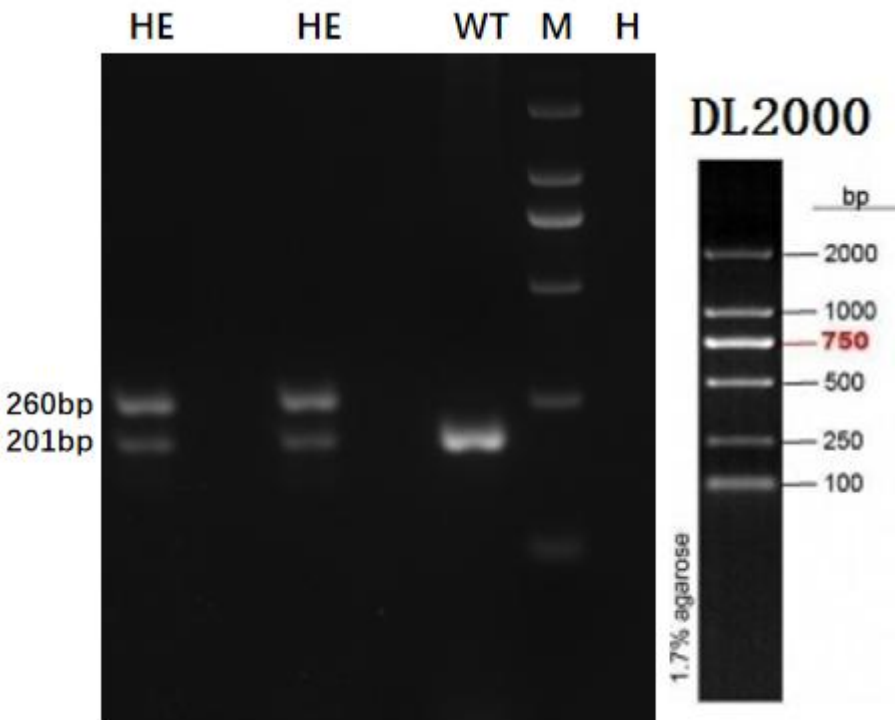


→ : primer location

Primers

Primer	Sequence (5' → 3')	Primer type
P1	TCAAAGAGCCAATCAGGAAGTT	Forward
P2	GGGGCAGATGACAGAAGTTGGATT	Reverse

Expected results

Results	
Genotype	Wild type: P1P2 =201 bp Heterozygote: P1P2 =201 bp and 260 bp Homozygote: P1P2 =260 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	