

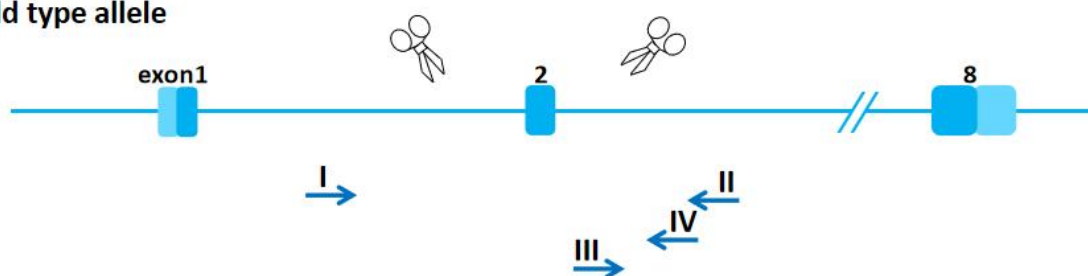
Zdhhc18-K0 Genotyping Protocol



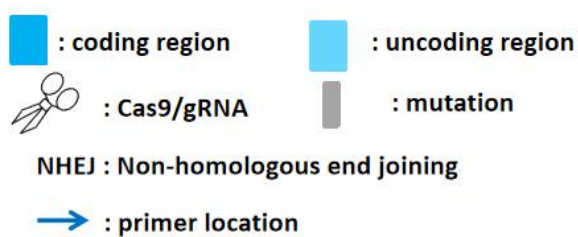
Common Name	Zdhhc18-K0	Cat. NO.	NM-K0-234985
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy

Wild type allele



knockout allele

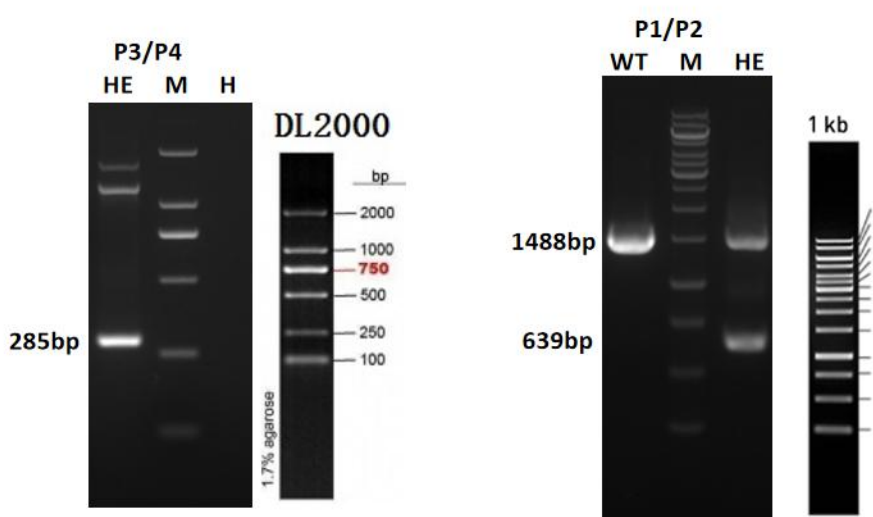


Primers

Primer	Sequence (5' → 3')	Primer type
P1	TTGGACCTTCTGAGTGTGCG	Forward
P2	GTTTAGCCAAAGGTGGGGGA	Reverse

P3	GCAGGCAAATATGTAAGCAAGA	Forward
P4	CTGGCCTCCAACCTCACAG	Reverse

Expected results

Results	 P3/P4 HE M H 285bp DL2000 bp 2000 1000 750 500 250 100 1.7% agarose P1/P2 WT M H 1488bp 639bp 1 kb bp 10000 8000 6000 5000 4000 3000 2500 2000 1500 1000 750 500 250
Genotype	Knockout type: -849 bp Wild type: P1P2 =1488 bp; P3P4=285 bp Heterozygote: P1P2 =1488 bp and 639 bp; P3P4=285 bp Homozygote: P1P2 =639 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 System	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	
	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	