

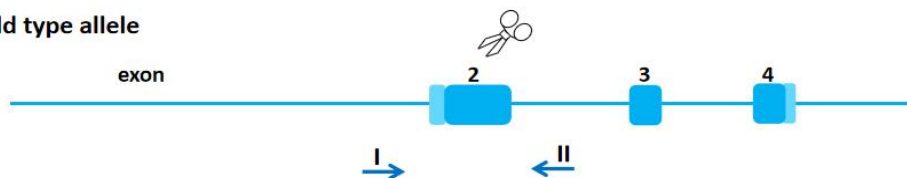
Cebpz^{os}-K0 Genotyping Protocol



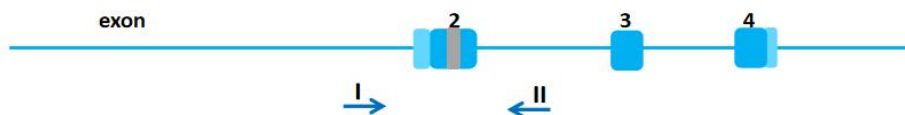
Common Name	Cebpz0s-K0	Cat. NO.	NM-K0-232131
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy

Wild type allele



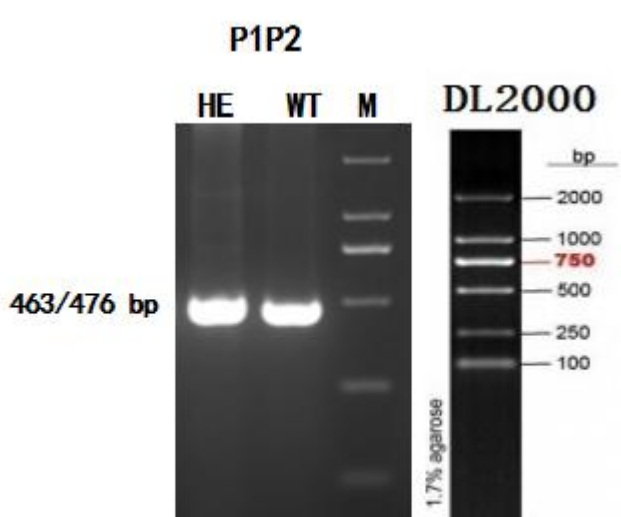
knockout allele



Primers

Primer	Sequence (5' → 3')	Primer type
P1	CCTTAGGCATGGAGATCGGAA	Forward
P2	AGCCAGAATAGGGACGTCTG	Reverse
Seq primer	CCTTAGGCATGGAGATCGGAA	Sequencing primer

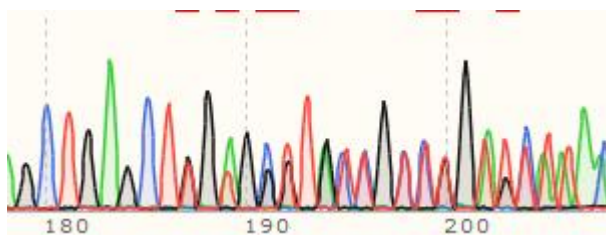
Expected results

Results	P1P2 
Genotype	Knockout type: -13bp Wild type: P1P2 =476 bp Heterozygote: P1P2 =476 bp and 463 bp; Homozygote: P1P2 =463 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

The PCR product should be sequencing. Sequencing peak data is for identifying homozygous, heterozygous and wild type mice.

Heterozygous (there are two peaks on mutant site) :



Reaction &Cycling

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

	Genomic DNA			1.0
	Total			20
	2×Taq Plus Master Mix from Vazyme (Code Number: P222-1)			
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	