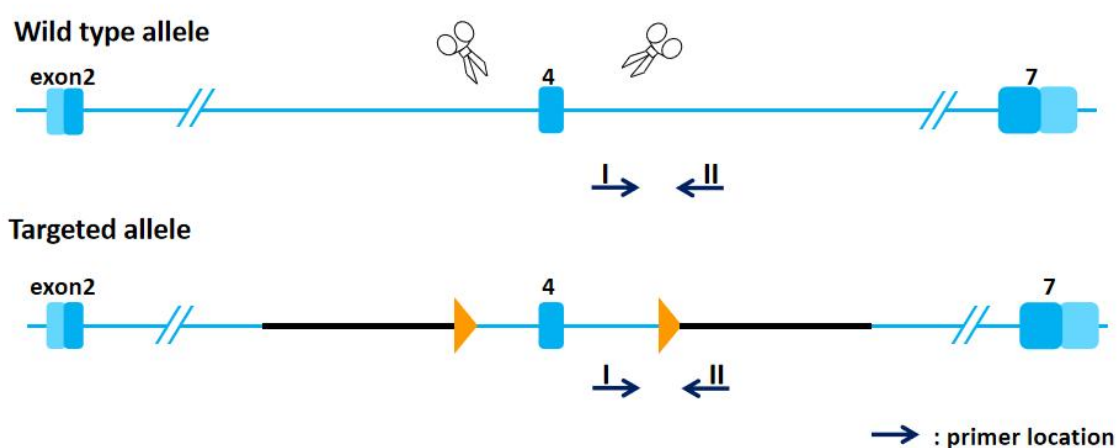


Atf1-Flox Genotyping Protocol



Common Name	Atf1-Flox	Cat. NO.	NM-CKO-240651
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy



Primers

Primer	Sequence (5'→3')	Primer type
P1	TCCCACGTGATAGACACACA	Forward
P2	GTCAGCAACAGTTTAAAAAGATGAA	Reverse

Expected results

Results	Agarose gel electrophoresis image showing PCR results. Lanes are labeled HE, HE, WT, M, H. Molecular weight markers are shown on the right: 2000, 1000, 750 (red), 500, 250, 100 bp. Specific bands are labeled 374bp and 309bp on the left.
Genotype	Wild type: P1P2 =309 bp; Heterozygote: P1P2 =374 bp and 309 bp; Homozygote: P1P2 =374 bp

Reaction &Cycling

	Reaction Component	Volume (μ l)
PCR Reaction System	ddH ₂ O	8.0
	2 × Rapid Taq Master Mix	10.0
	P1(10 pmol/ μ l)	0.5
	P2(10 pmol/ μ l)	0.5
	Genomic DNA	1.0
	Total	20

	<i>2 × Rapid Taq Master Mix from Vazyme(Code Number: P222-01)</i>			
<i>Cycling Reaction</i>	<i>Step</i>	<i>Temp</i>	<i>Time</i>	<i>Note</i>
	<i>1</i>	<i>95°C</i>	<i>5 min</i>	
	<i>2</i>	<i>95°C</i>	<i>20 sec</i>	
	<i>3</i>	<i>60°C</i>	<i>20 sec</i>	
	<i>4</i>	<i>72°C</i>	<i>20 sec</i>	<i>35 repeats to 2</i>
	<i>5</i>	<i>72°C</i>	<i>5 min</i>	
	<i>6</i>	<i>12°C</i>	<i>Hold</i>	