

# Zfp449 Cas9-KO Strategy

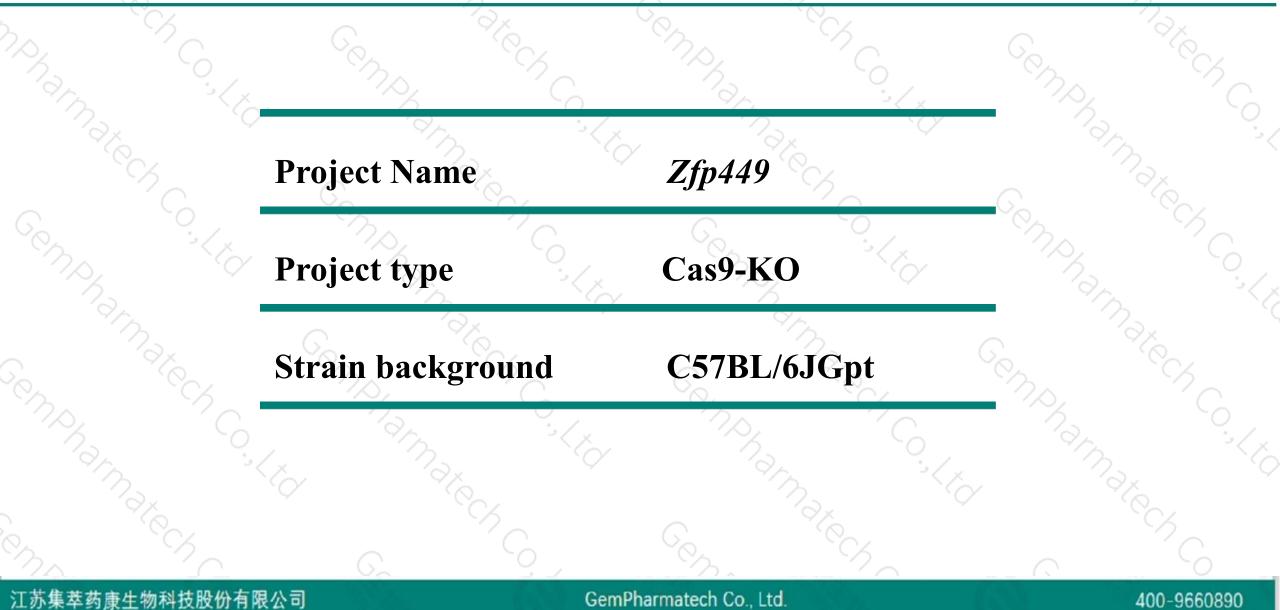
**Designer: Zihe Cui** 

**Reviewer: Xueting Zhang** 

Design Date: 2020-10-10

### **Project Overview**

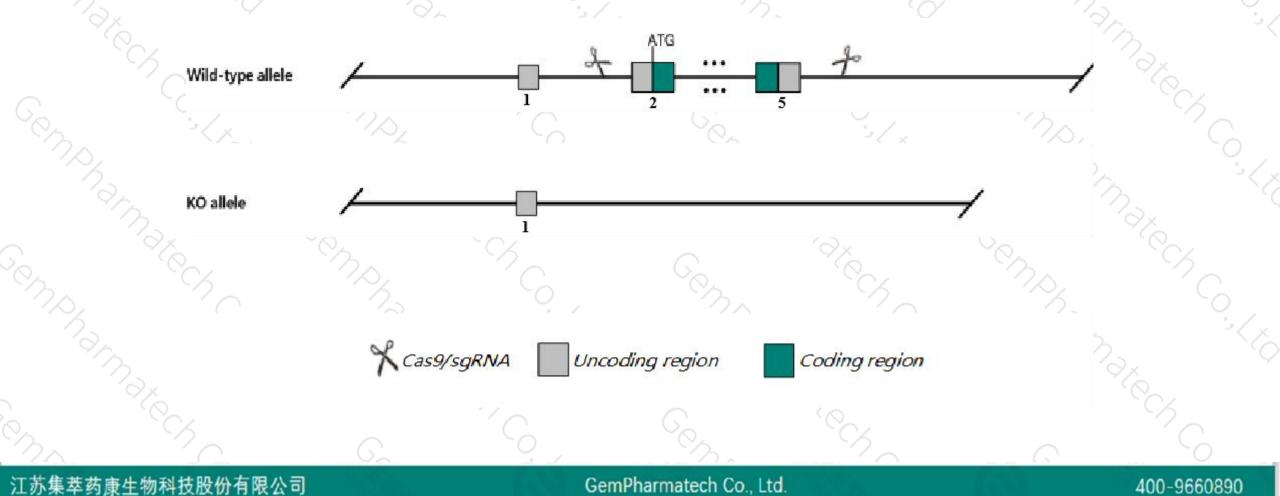




### **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the Zfp449 gene. The schematic diagram is as follows:





> The Zfp449 gene has 1 transcript. According to the structure of Zfp449 gene, exon2-exon5 of Zfp449-201(ENSMUST00000101560.3) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.

> In this project we use CRISPR/Cas9 technology to modify Zfp449 gene. The brief process is as follows: CRISPR/Cas9 system were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- > According to the existing MGI data, mice homozygous for a knock-out allele exhibit normal skeletal growth and cartilage homeostasis.
- The Zfp449 gene is located on the ChrX. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
   This strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes,all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Notice

## **Gene information (NCBI)**



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#### Zfp449 zinc finger protein 449 [ Mus musculus (house mouse) ]

Gene ID: 78619, u	updated on	25-Sep-2020
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#### Summary

Official Symbol	Zfp449 provided by MGI						
Official Full Name	zinc finger protein 449 provided by MGI						
Primary source	MGI:MGI:1925869						
See related	ed Ensembl:ENSMUSG0000073176						
Gene type	protein coding						
RefSeq status	VALIDATED						
Organism	Mus musculus						
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;						
	Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus						
Also known as	9530049C15Rik						
Expression	<ul> <li>Broad expression in limb E14.5 (RPKM 2.2), bladder adult (RPKM 1.6) and 21 other tissues See more</li> </ul>						
Orthologs	human all						

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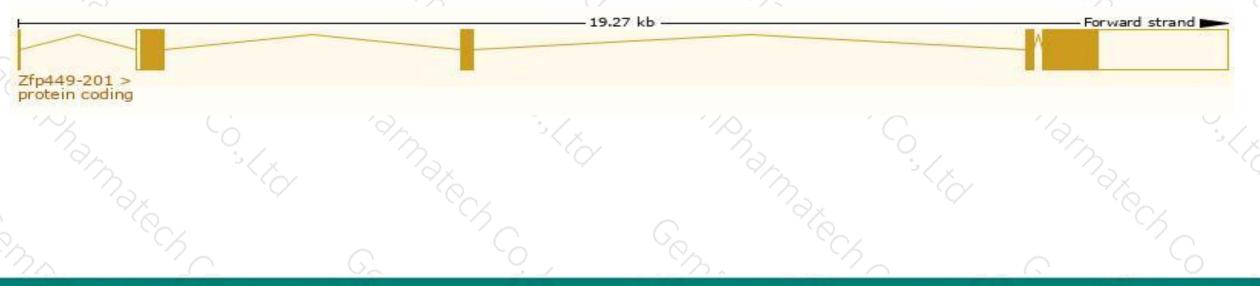
### **Transcript information (Ensembl)**



The gene has 1 transcript, and the transcript is shown below:

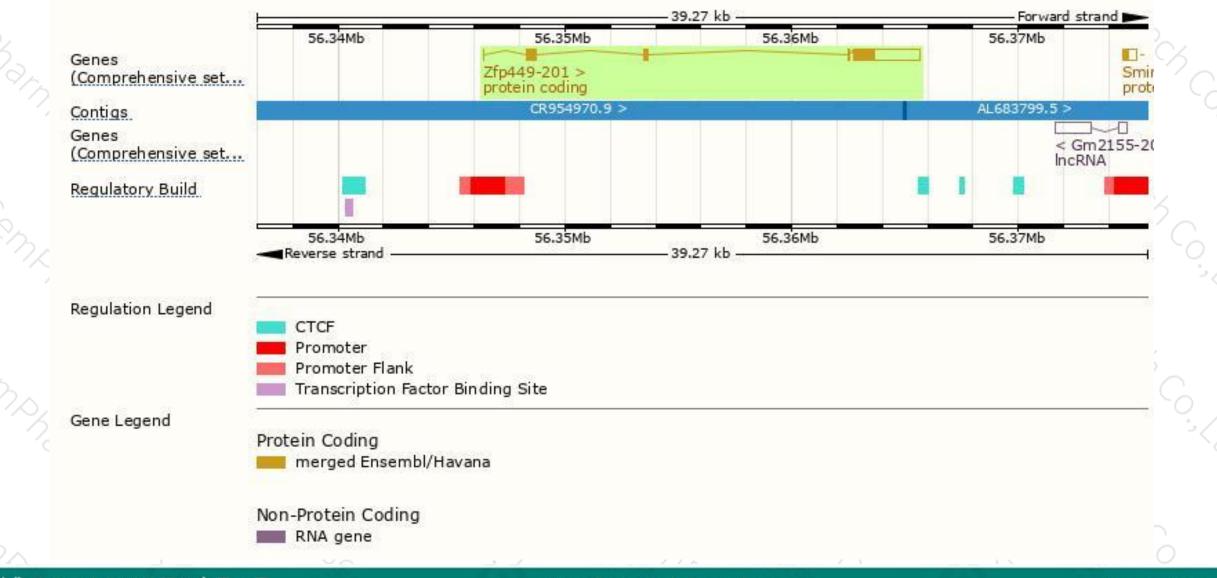
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Zfp449-201	ENSMUST00000101560.3	3738	<u>518aa</u>	Protein coding	CCD530143	<u>Q8CB76</u>	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of *Zfp449-201* transcript, the transcription is shown below:



### **Genomic location distribution**





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### **Protein domain**



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3.	ENSMUSP00000099 MobiDB lite Low complexity (Seg) Superfamily	SSF47353			Zinc finger C2H2 su	narfamily	300
	SMART	SCAN domain			Zinc finger C2H2-t		
	Pfam	SCAN domain			Zinc finger C2H2-t		
	PROSITE profiles	SCAN domain	1		Zinc finger C2H2-t		
9	PROSITE patterns PANTHER	PTHR23226			Zinc finger C2H2-I	уре	<u> </u>
	Gene3D	PTHR23226:SF4			3.30.160.60	iii iii	
	CDD	SCAN domain su SCAN domain	perrantiny		5.50.160.60	0065	
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	Variant Legend	missense vari					
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If you have any questions, you are welcome to inquire. Tel: 400-9660890



