

# Mzf1 Cas9-CKO Strategy

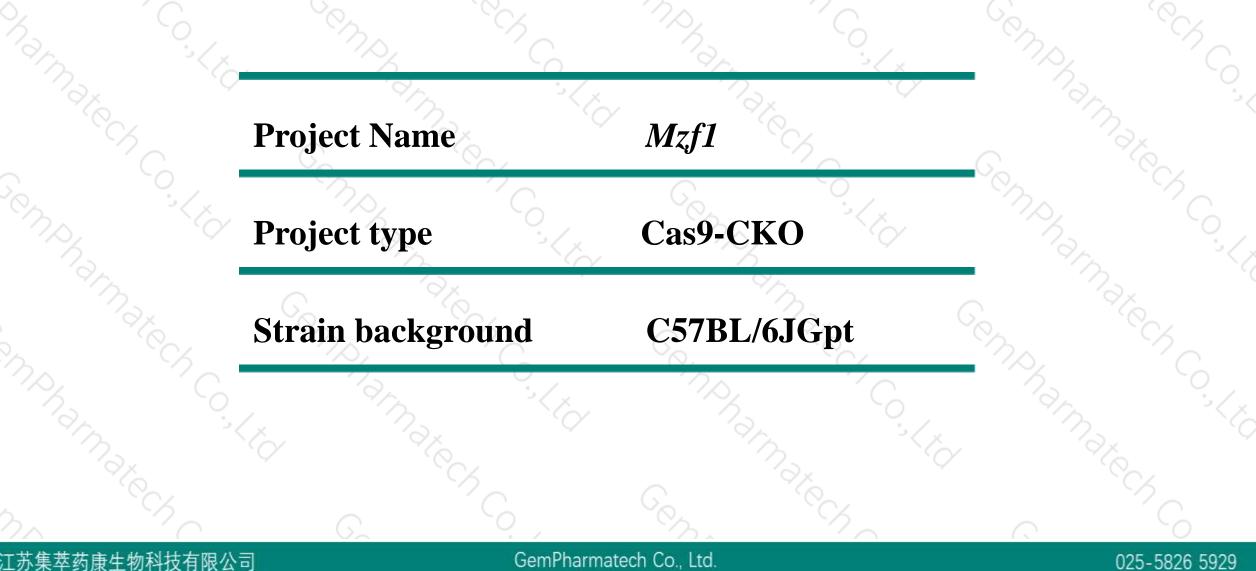
Designer: Reviewer:

**Design Date:** 

Jing Jin Yang Zeng 2018-9-25

# **Project Overview**





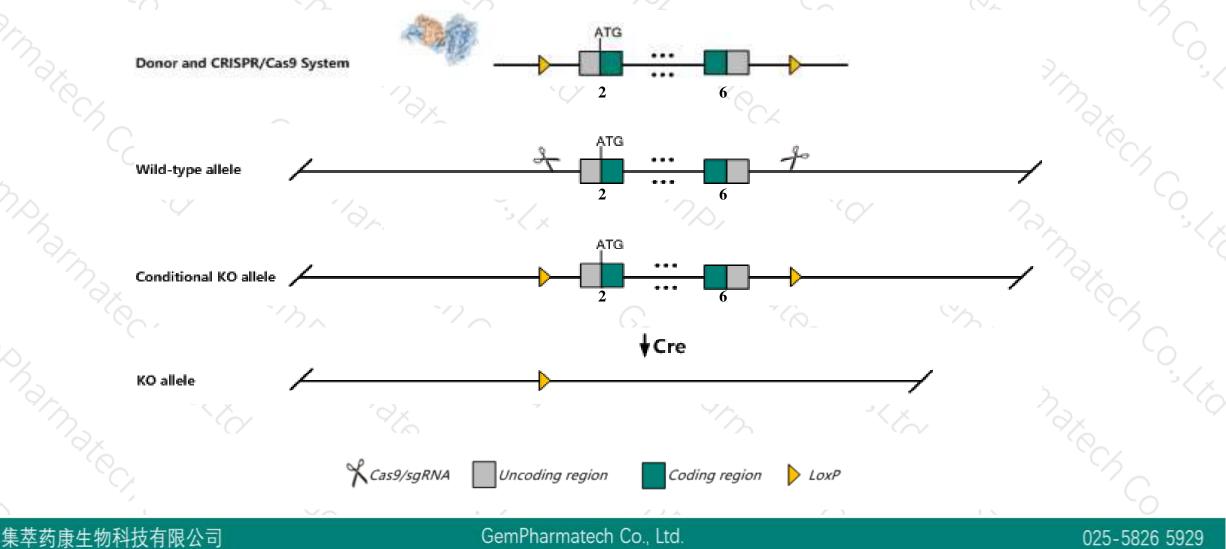
技有限公司 江苏集萃药

GemPharmatech Co., Ltd.

# **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Mzf1* gene. The schematic diagram is as follows:





- The *Mzf1* gene has 7 transcripts. According to the structure of *Mzf1* gene, exon2-exon6 of *Mzf1-205* (ENSMUST00000182515.7) 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 *Mzf1* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA and Donor 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.
- The flox mice was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

# Notice



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- According to the existing MGI data, Homozygous mutants are prone to late-onset (>2 yr) neoplasias characterized by infiltration, enlargement and disruption of the liver by monomorphic cells of myeloid origin; hemopoietic progenitors show increased ability to sustain long-term hemopoiesis.
- The Mzf1 is close to 5'UTR region of Chmp2a and Ube2m gene. Knockout the region may affect the function of Chmp2a and Ube2m gene.
- The *Mzf1* gene is located on the Chr7. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

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## **Gene information (NCBI)**



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### Mzf1 myeloid zinc finger 1 [ Mus musculus (house mouse) ]

Gene ID: 109889, updated on 12-Aug-2019

#### Summary

Official Symbol Mzf1 provided by MGI Official Full Name myeloid zinc finger 1 provided by MGI Primary source MGI:MGI:107457 See related Ensembl:ENSMUSG00000030380 Gene type protein coding **RefSeq status** VALIDATED Mus musculus Organism Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Lineage Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus Also known as Mzf2; Mzf-2; Zfp98; Znf42; Zfp121 Ubiquitous expression in adrenal adult (RPKM 1.8), frontal lobe adult (RPKM 1.7) and 27 other tissues See more Expression Orthologs human all

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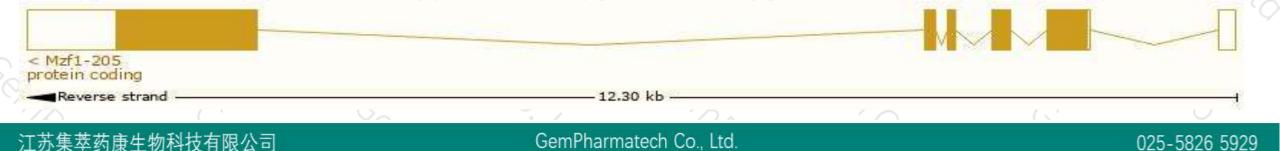
# **Transcript information (Ensembl)**



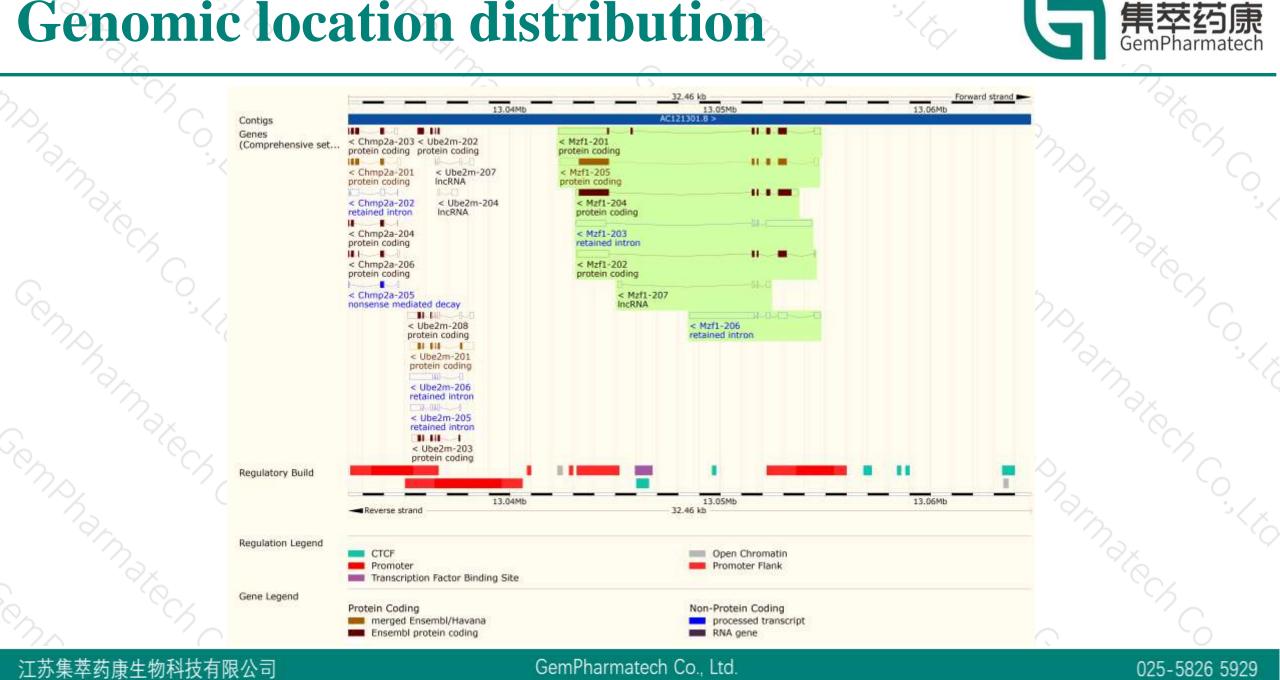
### The gene has 7 transcripts, all transcripts are shown below:

Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags		
ENSMUST00000182515.7	3340	<u>735aa</u>	Protein coding	CCDS20826	<u>A1L358</u>	TSL:1 GENCODE basic APPRIS P2		
ENSMUST0000069289.14	3597	320aaProtein coding814aaProtein coding		-	F6VC78	TSL:1 GENCODE basic		
ENSMUST00000182490.1	2872			-	<u>S4R1L6</u>	TSL:1 GENCODE basic APPRIS ALT2		
ENSMUST00000182087.1	2197	<u>190aa</u>	Protein coding	-	<u>S4R1C2</u>	TSL:1 GENCODE basic		
ENSMUST00000210727.1	578	No protein	No protein Processed transcript		-	TSL:5		
ENSMUST00000183176.1	4015	No protein	Retained intron	-	-	TSL:1		
ENSMUST00000182293.7	3749	No protein	protein Retained intron		-	TSL:1		
	ENSMUST00000182515.7 ENSMUST0000069289.14 ENSMUST00000182490.1 ENSMUST00000182087.1 ENSMUST00000210727.1 ENSMUST00000183176.1	ENSMUST00000182515.7 3340   ENSMUST0000069289.14 3597   ENSMUST00000182490.1 2872   ENSMUST00000182087.1 2197   ENSMUST00000182087.1 578   ENSMUST00000183176.1 4015	ENSMUST00000182515.7 3340 735aa   ENSMUST0000069289.14 3597 320aa   ENSMUST0000182490.1 2872 814aa   ENSMUST00000182087.1 2197 190aa   ENSMUST0000018210727.1 578 No protein   ENSMUST00000183176.1 4015 No protein	ENSMUST00000182515.73340735aaProtein codingENSMUST0000069289.143597320aaProtein codingENSMUST00000182490.12872814aaProtein codingENSMUST00000182087.12197190aaProtein codingENSMUST00000210727.1578No proteinProcessed transcriptENSMUST00000183176.14015No proteinRetained intron	ENSMUST00000182515.73340735aaProtein codingCCDS20826ENSMUST0000069289.143597320aaProtein coding-ENSMUST0000182490.12872814aaProtein coding-ENSMUST0000182087.12197190aaProtein coding-ENSMUST00000210727.1578No proteinProcessed transcript-ENSMUST0000183176.14015No proteinRetained intron-	ENSMUST00000182515.73340735aaProtein codingCCDS20826A1L358ENSMUST0000069289.143597320aaProtein codingF6VC78ENSMUST0000182490.12872814aaProtein codingS4R1L6ENSMUST0000182087.12197190aaProtein codingS4R1C2ENSMUST0000210727.1578No proteinProcessed transcript-ENSMUST0000183176.14015No proteinRetained intron-		

The strategy is based on the design of *Mzf1-205* transcript, The transcription is shown below



### **Genomic location distribution**



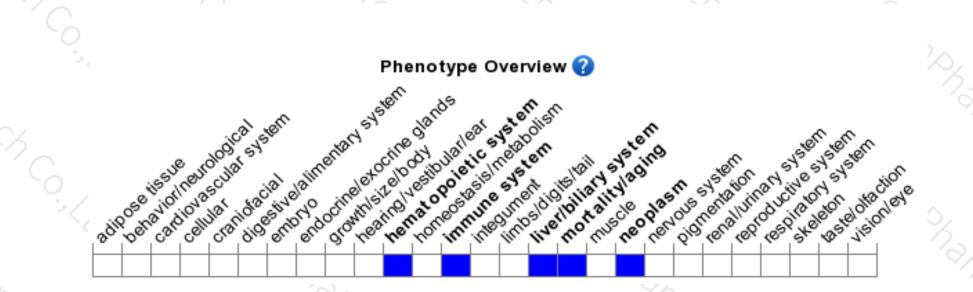
### **Protein domain**



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### Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Homozygous mutants are prone to late-onset (>2 yr) neoplasias characterized by infiltration, enlargement and disruption of the liver by monomorphic cells of myeloid origin; hemopoietic progenitors show increased ability to sustain long-term hemopoiesis.



If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



