

# Serpina10 Cas9-CKO Strategy

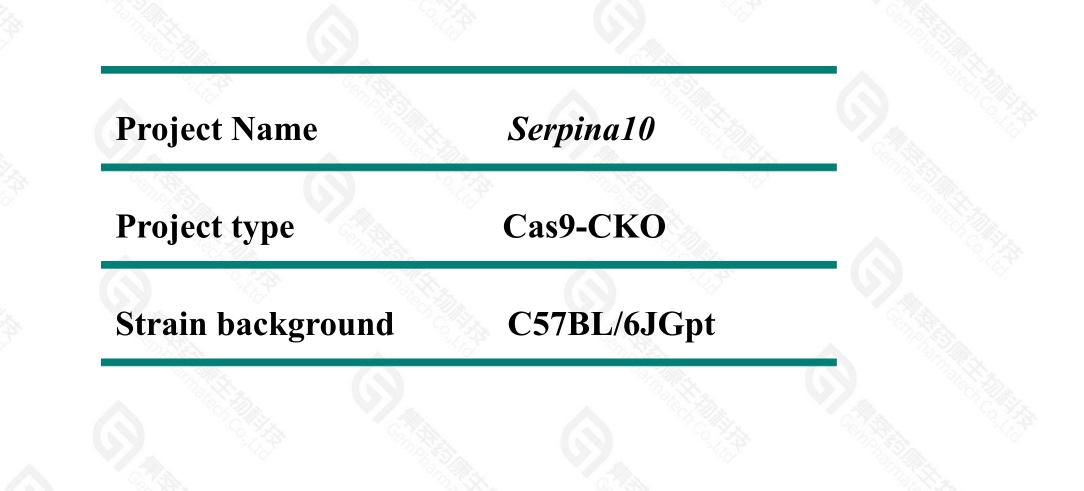
**Designer: Rui Xiong** 

**Reviewer: Longyun Hu** 

**Design Date: 2021-6-26** 

# **Project Overview**



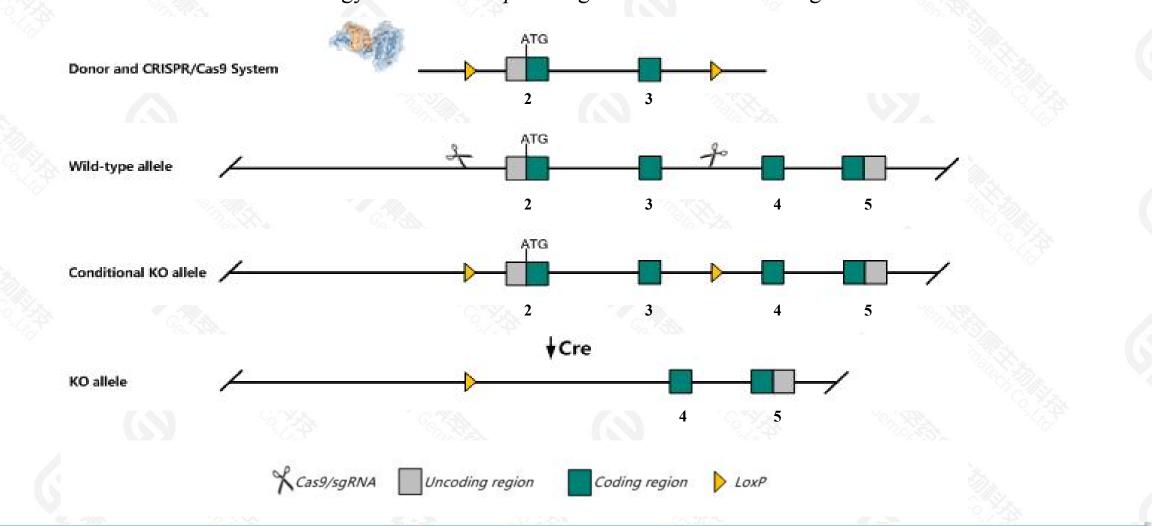


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# **Conditional Knockout strategy**

This model will use CRISPR/Cas9 technology to edit the *Serpina10* gene. The schematic diagram is as follows:



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### **Technical routes**



The Serpinal0 gene has 2 transcripts. According to the structure of Serpinal0 gene, exon2-exon3 of Serpinal0-201(ENSMUST00000044231.12) transcript is recommended as the knockout region. The region contains start codon ATG.Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Serpina10* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor 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.



- > According to the existing MGI data,mice homozygous for a knock-out allele display a reduced survival rate, enhanced thrombosis after ferric chloride-induced carotid artery injury, and increased mortality from pulmonary thromboembolism following collagen/epinephrine infusion.
- The Serpina10 gene is located on the Chr12. 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.

# Gene information (NCBI)



### Serpina10 serine (or cysteine) peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 10 [Mus musculus (house mouse)]

Gene ID: 217847, updated on 25-Sep-2020

### Summary \$ ? Official Symbol Serpina10 provided by MGI Official Full Name serine (or cysteine) peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 10 provided by MGI Primary source MGI:MGI:2667725 See related Ensembl:ENSMUSG00000061947 Gene type protein coding RefSeq status REVIEWED Organism Mus musculus Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus Also known as P. PZI, ZPI Summary The protein encoded by this gene is a member of the large serpin family of proteins, and is also known as serpin PZdependent protease inhibitor (ZPI or PZI). This protein is thought to play an important role in the regulation of coagulation. It directly inhibits factor XIa, and also inhibits factor Xa in the presence of calcium, phospholipids, and protein Z (PZ). Deficiencies in this gene lead to an increase in thrombosis. Alternative splicing results in multiple transcript variants that encode multiple protein isoforms. [provided by RefSeq, Aug 2014] Expression Biased expression in liver E18 (RPKM 38.8), liver adult (RPKM 34.0) and 3 other tissuesSee more Orthologs human all

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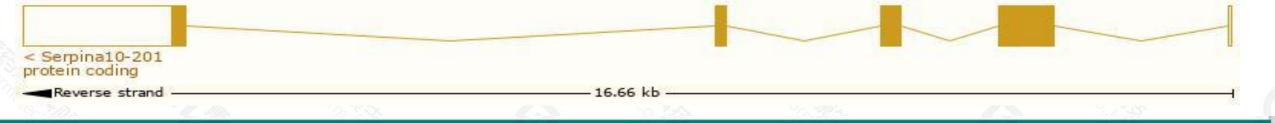
# **Transcript information (Ensembl)**



The gene has 2 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags		
Serpina10-201	ENSMUST00000044231.12	3498	<u>448aa</u>	Protein coding	CCD526134		TSL:1, GENCODE basic, APPRIS P1,		
Serpina10-202	ENSMUST00000121625.2	1424	<u>394aa</u>	Protein coding	CCDS79150		TSL:1 , GENCODE basic ,		

The strategy is based on the design of *Serpina10-201* transcript, the transcription is shown below:

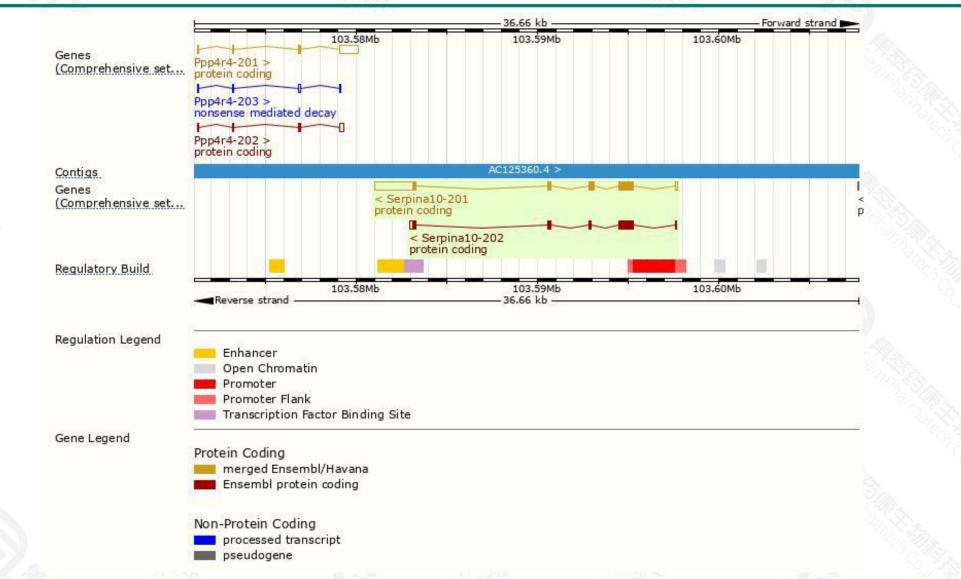


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### **Genomic location distribution**





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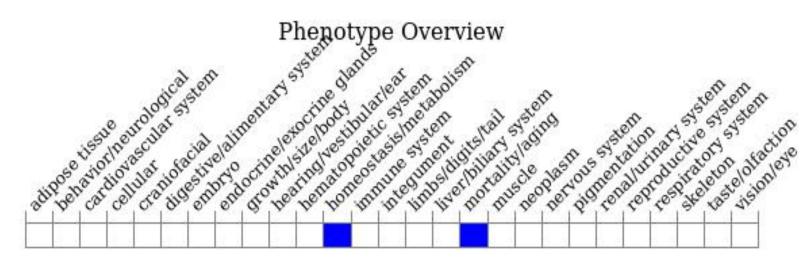
# **Protein domain**



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Superfamily	Serpin superfamily									
SMART	Serpi	n domain								10
Prints	PR00780					-				
Pfam.	Serpin	domain								
PANTHER	PTHR11461:SF19	1								
Cana 2D	Serpin family									-
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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,mice homozygous for a knock-out allele display a reduced survival rate, enhanced thrombosis after ferric chloride-induced carotid artery injury, and increased mortality from pulmonary thromboembolism following collagen/epinephrine infusion.

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



