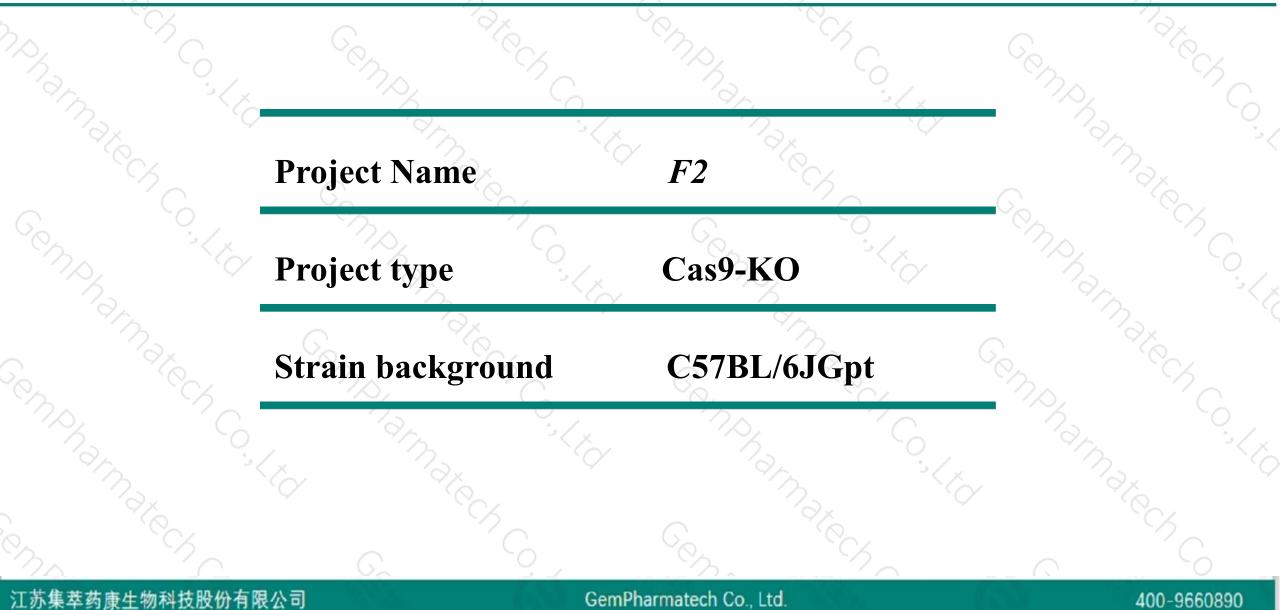


# F2 Cas9-KO Strategy

Designer: Reviewer: Design Date: Huimin Su Ruirui Zhang 2019/8/30

### **Project Overview**

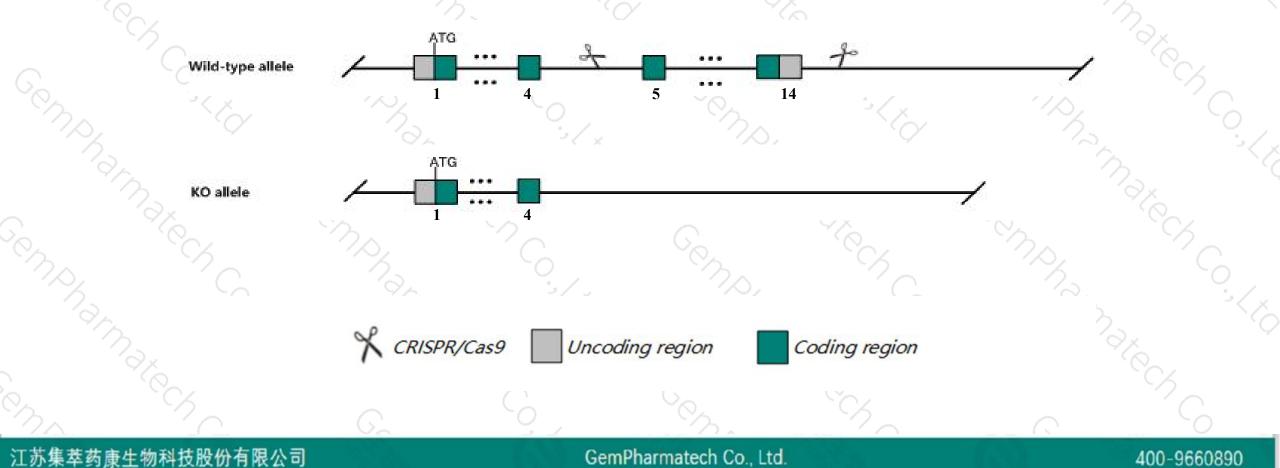




## **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the F2 gene. The schematic diagram is as follows:





- The F2 gene has 3 transcripts. According to the structure of F2 gene, exon5-exon14 of F2-201 (ENSMUST0000028681.14) transcript is recommended as the knockout region. The region contains 1538bp coding sequenc Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify F2 gene. The brief process is as follows: CRISPR/Cas9 system we

400-9660890

- According to the existing MGI data, Homozygotes for targeted null mutations exhibit defects in yolk sac vasculature, internal bleeding, tissue necrosis, and die in mid- to late-gestation, or rarely, a few days after birth.
- The F2 gene is located on the Chr2. 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)



\$ ?

#### F2 coagulation factor II [ Mus musculus (house mouse) ]

Gene ID: 14061, updated on 12-Aug-2019

Summary

Official Symbol F2 provided by MGI Official Full Name coagulation factor II provided by MGI Primary source MGI:MGI:88380 See related Ensembl:ENSMUSG00000027249 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 Cf2: FII: Cf-2 Summary This gene encodes a vitamin K-dependent glycoprotein coagulation factor that plays an important role in the process of blood coagulation and hemostasis. The encoded protein is an inactive zymogen that undergoes enzymatic cleavage by the coagulation factor Xa to form an active serine protease that converts soluble fibrinogen to insoluble fibrin clot. Most of the mice lacking the encoded protein die at an embryonic stage due to defects in yolk sac vasculature, while the rare nenonates succumb to hemorrhage on the first postnatal day. [provided by RefSeq, Apr 2015] Expression Biased expression in liver adult (RPKM 695.9), liver E18 (RPKM 621.7) and 3 other tissues See more Orthologs human all

#### 江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.

#### 400-9660890

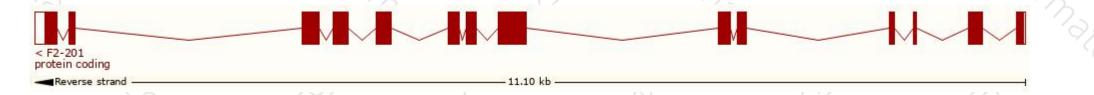
# **Transcript information (Ensembl)**



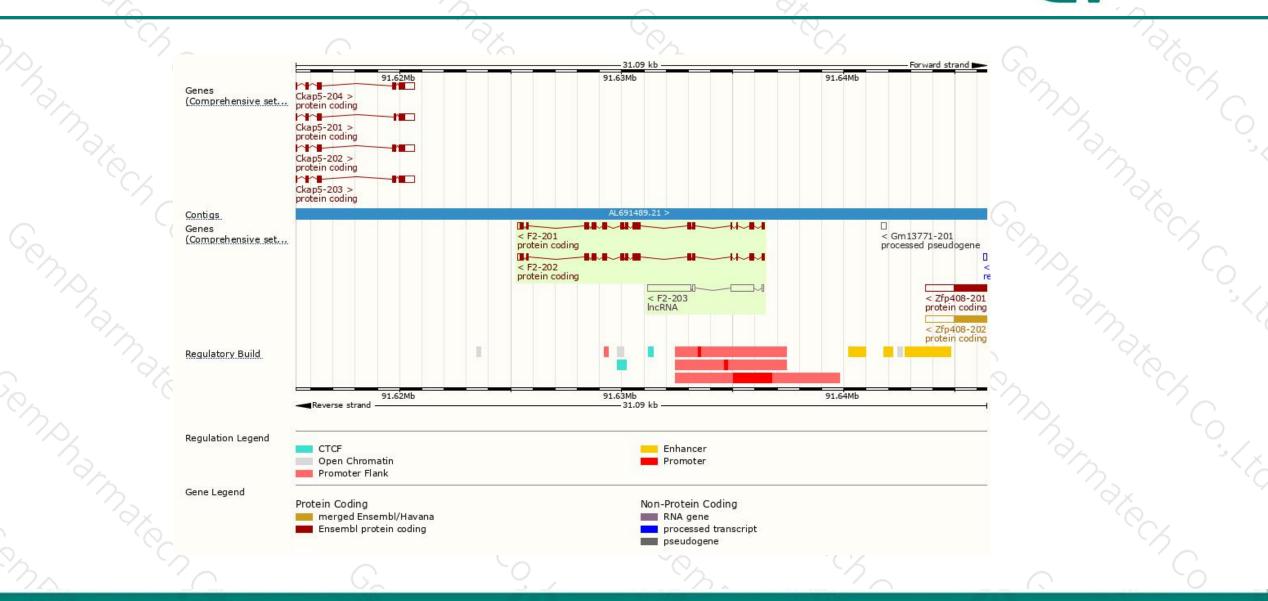
The gene has 3 transcripts, all transcripts are shown below:

Name 🖕	Transcript ID 🖕	bp 🍦	Protein 🛔	Biotype 🍦	CCDS 🍦	UniProt 🖕	Flags 🍦		
F2-201	ENSMUST0000028681.14	1988	<u>618aa</u>	Protein coding	<u>CCDS16434</u> &	P19221 & Q3TJ94 &	TSL:1	GENCODE basic	APPRIS P2
F2-202	ENSMUST00000111335.1	1969	<u>617aa</u>	Protein coding		<u>H7BX99</u> &	TSL:5	GENCODE basic	APPRIS ALT2
F2-203	ENSMUST00000153182.1	3167	No protein	IncRNA	8	β.	TSL:2		

The strategy is based on the design of *F2-201* transcript, The transcription is shown below



## **Genomic location distribution**



#### 江苏集萃药康生物科技股份有限公司

### GemPharmatech Co., Ltd.

#### 400-9660890

集举药康 GemPharmatech

# **Protein domain**



	$\sim$			
	ENSMUSP00000028 SIFTS import Low complexity (Seg)	_		°C/
	Cleavage site (Sign	and the second se		
$(\mathcal{D})$	Superfamily	Gamma-carboxyglutamic acid-rich (GLA) domain superfamily	Peptidase S1, PA clan	
<sup>C</sup>	SHART.	Kringle-like fold		
	SMART	Gamma-carboxyglutamic acid-rich (GLA) domain	Serine proteases, trypsin domain	$\mathcal{D}_{\mathcal{A}}$
	20208	Kringle		
	Prints	Gamma-carboxyglutamic acid-rich (GLA) domain	Prothrombin/thrombin	
	Pfam	PR00018	Peptidase S1A, chymotrypsin family	
	Pidm.	Kringle	Thrombin light chain	
		Gamma-carboxyglutamic acid-rich (GLA) domain	Serine proteases, trypsin domain	
	PROSITE profiles	Kringle	Serine proteases, trypsin domain	· '31
1	PROCITE anthread	Gamma-carboxyglutamic acid-rich (GLA) domain		
	PROSITE patterns	Kringle, conserved site	Serine proteases	* 12J
	PIRSF	Gamma-carboxyglutamic acid-rich (GLA) domain	Serine proteases, trypsin family, histidine active site	
		Prothrombin/thrombin		
	PANTHER	PTHR24254		170
	C 3D	Prothrombin/thrombin		
X	Gene3D	Kringle superfamily	2.40.10.10	
	CDD		Thrombin light chain domain superfamily	2
~ 1	000	Kringle	Serine proteases, trypsin domain	
	All sequence SNPs/i	Sequence variants (dbSNP and all other sources)	15 21 15 17 1 T	9 Z.
	Variant Legend	ere a a a		
		missense variant	synonymous variant	
	Scale bar	<b>0</b> 60 120 180 240	300 360 420 480 540 618	
	( )		$ (\Delta ) $	

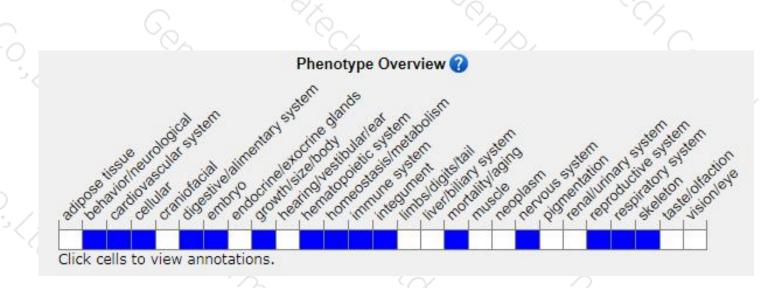
### 江苏集萃药康生物科技股份有限公司

### GemPharmatech Co., Ltd.

### 400-9660890

# 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, Homozygotes for targeted null mutations exhibit defects in yolk sac vasculature, internal bleeding, tissue necrosis, and die in mid- to late-gestation, or rarely, a few days after birth.



If you have any questions, you are welcome to inquire. Tel: 400-9660890



