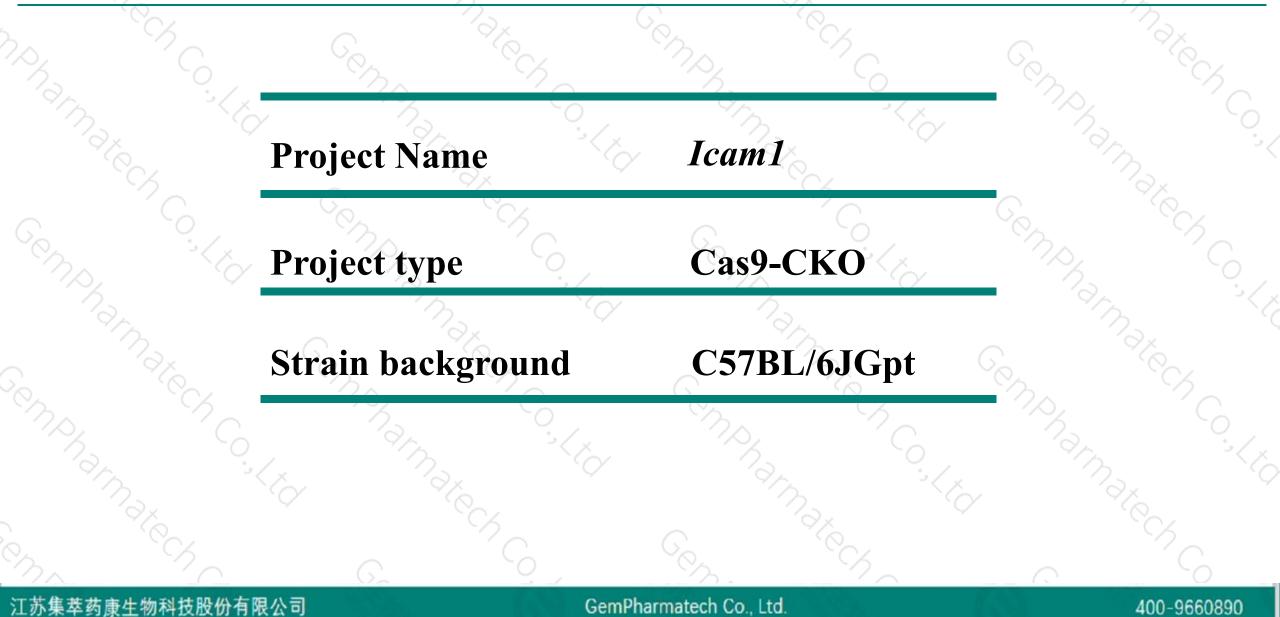
# Icam1 Cas9-CKO Strategy

Designer: Design Date: Jinling Wang 2019-7-26

### **Project Overview**

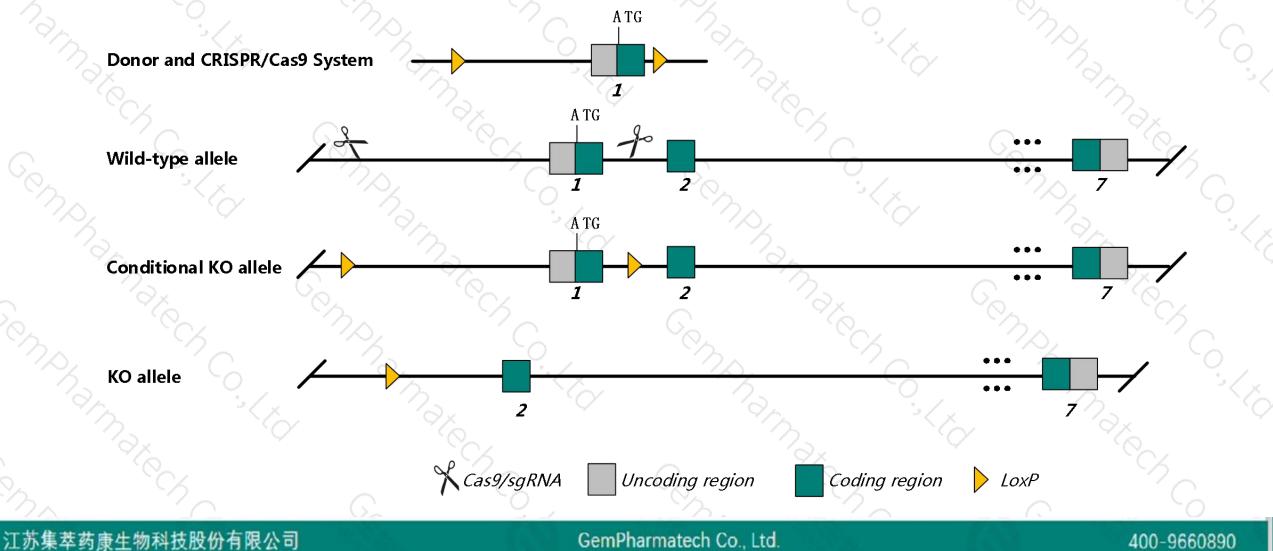




### **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Icam1* gene. The schematic diagram is as follows:





- The *Icam1* gene has 2 transcripts. According to the structure of *Icam1* gene, the predicted promoter region and exon1 of *Icam1*-201 (ENSMUST0000086399.5) transcript is recommended as the knockout region. The region contains the predicted promoter sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Icam1* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.



- According to the existing MGI data: Homozygous mutation of this gene results in impaired inflammatory and immune responses.
- The *Icam1* gene is located on the Chr9. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

### Gene information (NCBI)



8 ?

#### Icam1 intercellular adhesion molecule 1 [ Mus musculus (house mouse) ]

Gene ID: 15894, updated on 8-Dec-2018

#### Summary

 Official Symbol
 Icam1 provided by MGI

 Official Full Name
 intercellular adhesion molecule 1 provided by MGI

 Primary source
 MGI:MGI:96392

 See related
 Ensembl:ENSMUSG00000037405

 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
 CD54; Ly-47; Icam-1; MALA-2

 Summary
 This gene encodes an integral membrane protein that binds leukocyte adhesion protein LFA-1. It participates in the innate immune response. [provided by RefSeq, Aug 2016]

 Expression
 Biased expression in lung adult (RPKM 157.0), spleen adult (RPKM 29.7) and 6 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 2 transcripts, and all transcripts are shown below:

Name 🔺	Transcript ID	bp 🍦	Protein 🛊	Biotype 🝦	CCDS 🖕	UniProt 🖕	Flags			
Icam1-201	ENSMUST0000086399.5	2535	<u>537aa</u>	Protein coding	<u>CCDS22889</u> &	<u>P13597</u> & <u>Q3U8M7</u> &	TSL:1	GENCODE basic	APPRIS P1	
Icam1-202	ENSMUST00000215003.1	2203	No protein	Retained intron	5	-		TSL:2		

The strategy is based on the design of *Icam1*-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

集萃

GemPharmat

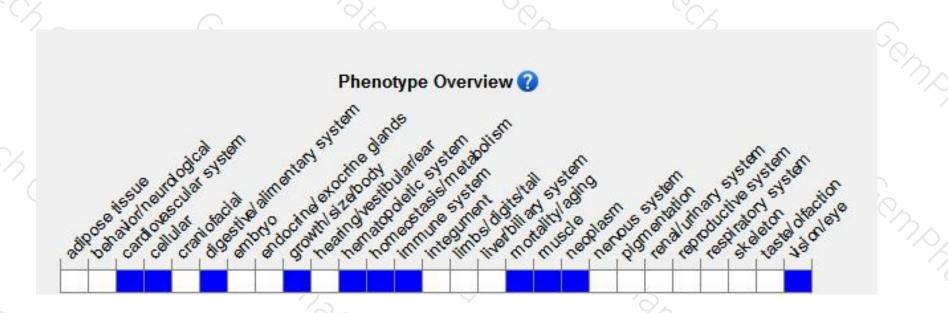
## **Protein domain**



ENSMUSP000	ne heli									-	20
Conserved Do Cleavage site	mains (Sign		_								
Superfamily d	PTER137	71 Immunoglobulin-like dom	ala suporfamily							1	
SMART domain	ns.	Immunoglobulin subtyp	e	dhesion molecule				-		~	
Pfam. domain.		Intercellular adhesion Intercellular adhesion mole	molecule/vascular cell ad ecule, N-terminal	hesion molecule, N-termin	nal	Immu	noglobulin-like damain				S
PROSITE profi	iles		-			Immu	noglobulin-like domain				
All sequence S	5NPs/i Sequenc	Immunoglobulin-Ilike fold e variants (dbSNP and a	il other sources)	$\hat{a} = \hat{a}_{1}$	(1, 1)	000 C		0	ĵ.	0	
Variant Legen		sense variant			synonymous variant						0
Scale bar	ō	60	120	180	240	300	360	420		537	
			1 Ce			1		Ŷ.		ALC CL	

### Mouse phenotype description(MGI)





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

Homozygous mutation of this gene results in impaired inflammatory and immune responses.

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



