

# Isg2012 Cas9-CKO Strategy

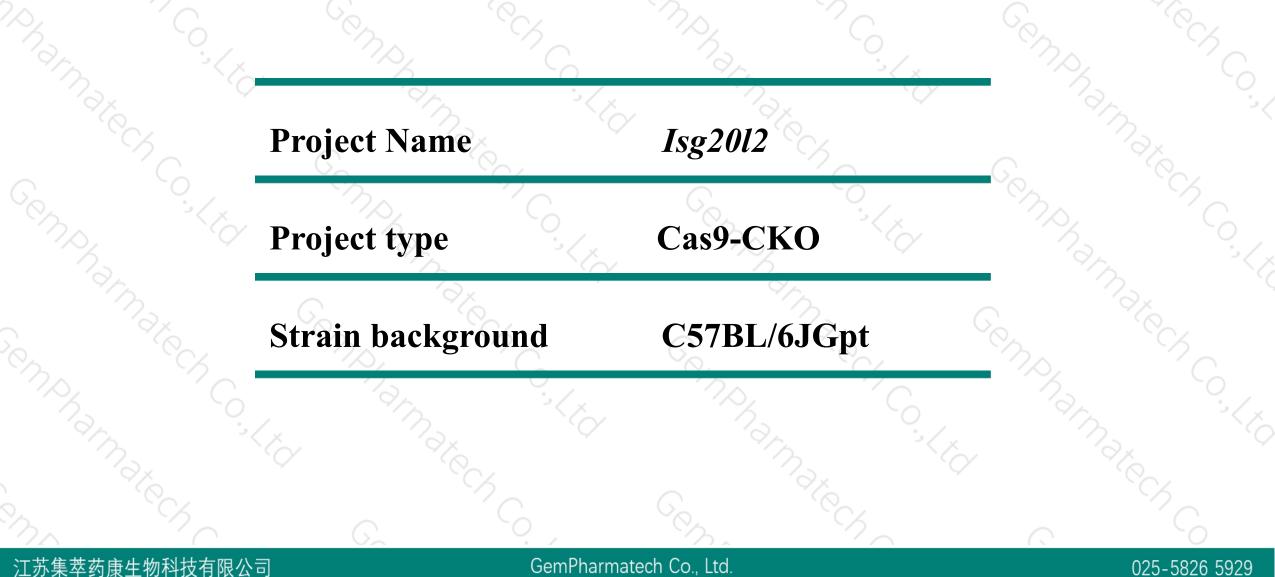
Designer: Jia Yu

**Reviewer:Xiaojing Li** 

**Design Date: 2020-9-25** 

## **Project Overview**





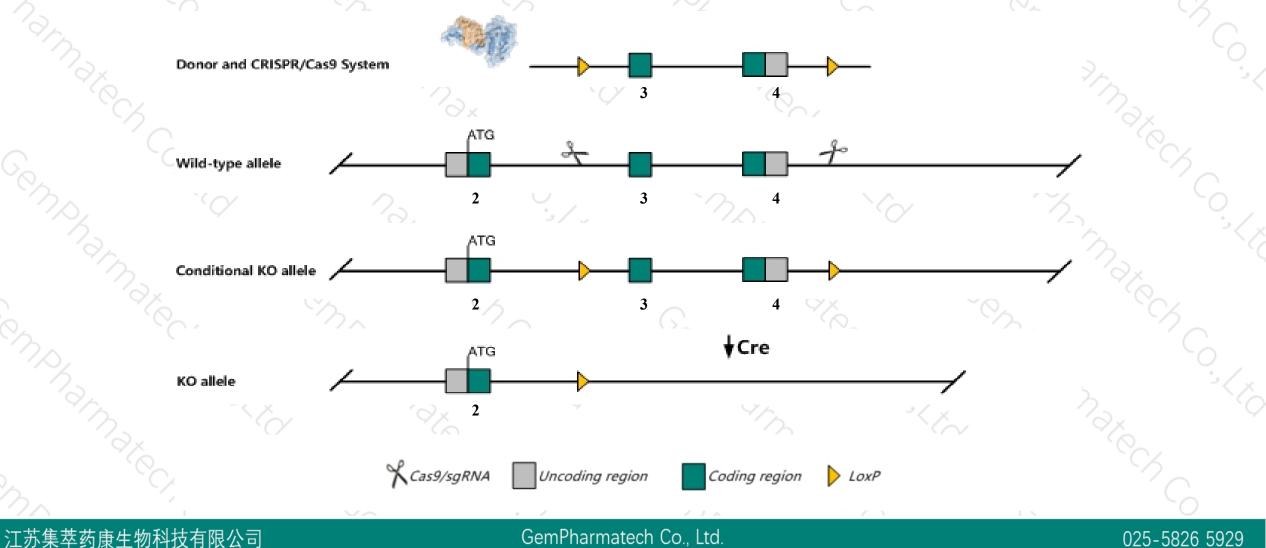
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## **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Isg20l2* gene. The schematic diagram is as follows:





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The *Isg2012* gene has 1 transcript. According to the structure of *Isg2012* gene, exon3-exon4 of *Isg2012*-201(ENSMUST00000055984.6) transcript is recommended as the knockout region. The region contains 315bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Isg20l2* 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.



- > The *Isg2012* gene is located on the Chr3. 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)**



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#### Isg20l2 interferon stimulated exonuclease gene 20-like 2 [Mus musculus (house mouse)]

Gene ID: 229504, updated on 13-Mar-2020

#### Summary

Official Symbol	Isg2012 provided by MGI
Official Symbol	isgzolz provided by MGI
Official Full Name	interferon stimulated exonuclease gene 20-like 2 provided by <u>MGI</u>
Primary source	MGI:MGI:2140076
See related	Ensembl:ENSMUSG0000048039
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	<u>Mus musculus</u>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;
	Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	4930429N24, C80587
Expression	Ubiquitous expression in testis adult (RPKM 26.3), CNS E11.5 (RPKM 14.8) and 28 other tissuesSee more
Orthologs	human all

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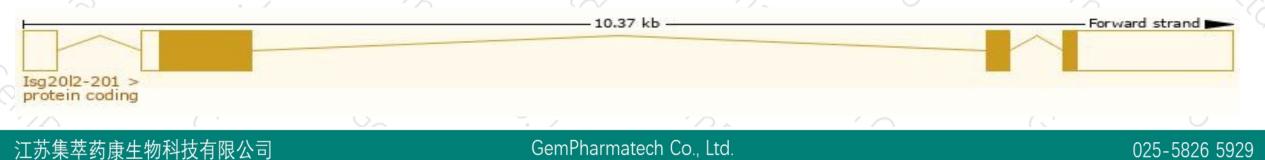
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The gene has 1 transcript, and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
lsg20l2-201	ENSMUST0000055984.6	2893	<u>368aa</u>	Protein coding	CCDS17459	A0A0R4J0R3	TSL:1 GENCODE basic AP	PRIS P1
°C/				<i>Q</i>				0

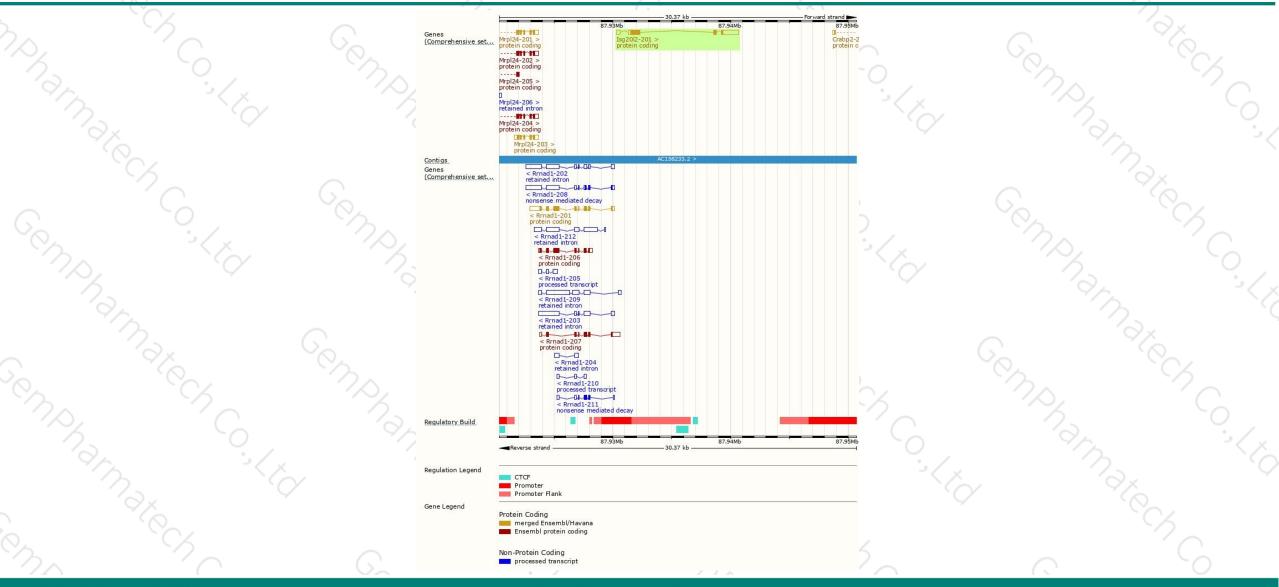
The strategy is based on the design of *Isg20l2-201* transcript, the transcription is shown below:



### **Genomic location distribution**



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## **Protein domain**



2	ENSMUSP00000059 MobiDB lite Low complexity (Seg) Superfamily				Ribonuclease H-like superfa	mily	× 6
	SMART				Exonuclease, RNase T/DNA	polymerase III	
	Pfam				Exonuclease, RNase T/DNA	polymerase III	-
	PANTHER	PTHR12801		1			
	Gene3D	Interferon-stimulated	20kDa exonuclease-li	ke 2	Ribonuclease H superfamily		
	CDD				ISG20, DEDDh 3'-5' exonuc	lease domain	
	All sequence SNPs/i	Sequence variants	(dbSNP and all othe	r sources)	HELEN L	10 TI 10	1.11
<,	Variant Legend	missense varia synonymous v stop retained	ant variant				0.4
	Scale bar	0 40	80	160	200 240	280 320	368
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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



