

# Sec14l2 Cas9-CKO Strategy

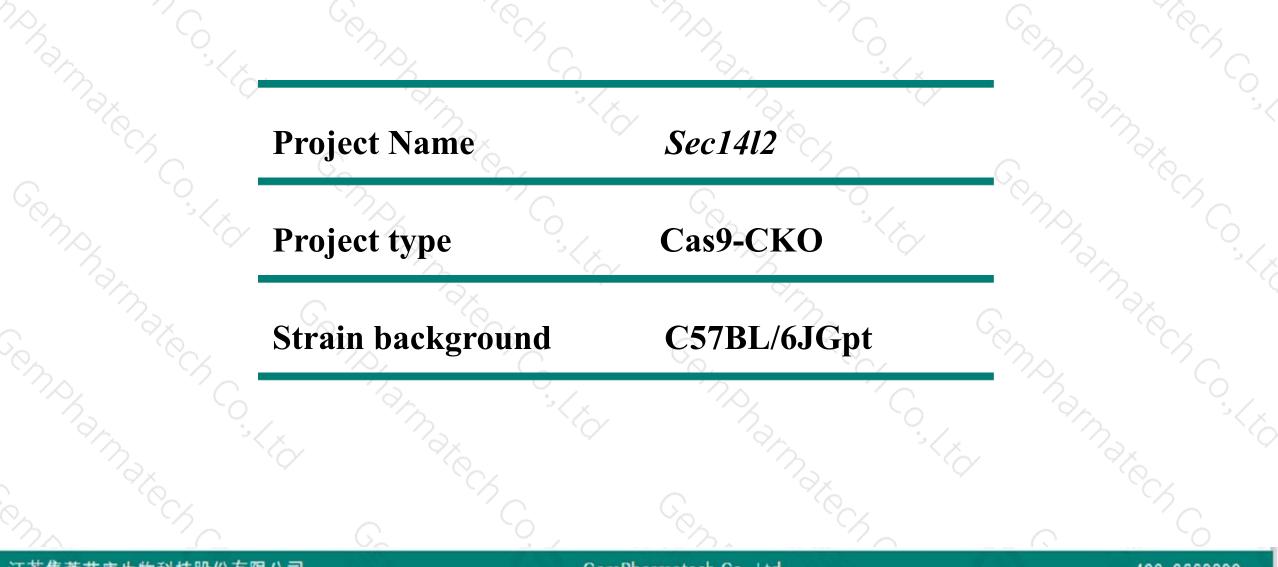
Designer: Reviewer:

**Design Date:** 

Daohua Xu Huimin Su 2020-1-20

## **Project Overview**





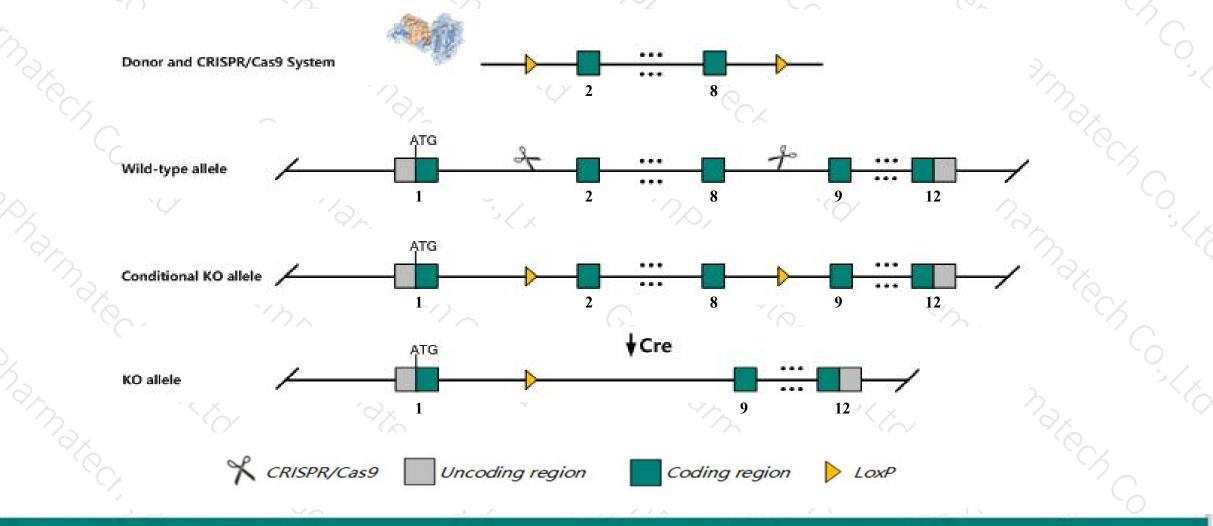
江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

## **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the Sec1412 gene. The schematic diagram is as follows:



江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.



The Sec14l2 gene has 6 transcripts. According to the structure of Sec14l2 gene, exon2-exon8 of Sec14l2-201 (ENSMUST0000003681.7) transcript is recommended as the knockout region. The region contains 610bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify Sec14l2 gene. The brief process is as follows:CRISPR/Cas9 system 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 and cell types.

## Notice



- According to the existing MGI data, Mice homozygous for a null allele exhibit decreased cholesterol synthesis and plasma levels under fasting conditions compared to wild-type mice.
- ➤The KO region contains functional region of the *Gm11955* gene.Knockout the region may affect the function of *Gm11955* gene.
- The Sec1412 gene is located on the Chr11. 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.

#### 江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.

## **Gene information (NCBI)**



\$ ?

#### Sec14I2 SEC14-like lipid binding 2 [Mus musculus (house mouse)]

Gene ID: 67815, updated on 31-Jan-2019

#### Summary

Official Symbol	Sec14I2 provided by MGI
Official Full Name	SEC14-like lipid binding 2 provided by MGI
Primary source	MGI:MGI:1915065
See related	Ensembl:ENSMUSG0000003585
Gene type	protein coding
<b>RefSeq status</b>	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1300013M05Rik, Spf, TAP
Expression	Biased expression in liver adult (RPKM 140.8), liver E18 (RPKM 27.5) 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.



The gene has 6 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sec1412-201	ENSMUST0000003681.7	2631	<u>403aa</u>	Protein coding	CCDS24377	<u>Q99J08</u>	TSL:1 GENCODE basic APPRIS P1
Sec14I2-202	ENSMUST00000123901.7	1624	No protein	IncRNA	-8	-	TSL:5
Sec14I2-203	ENSMUST00000132421.7	1229	No protein	IncRNA	22	-	TSL:5
Sec1412-204	ENSMUST00000133631.1	880	No protein	IncRNA	29	2	TSL:3
Sec14I2-205	ENSMUST00000136420.1	429	No protein	IncRNA	-	-	TSL:2
Sec1412-206	ENSMUST00000145173.1	423	No protein	IncRNA	-8	-	TSL:5

The strategy is based on the design of Sec1412-201 transcript, The transcription is shown below

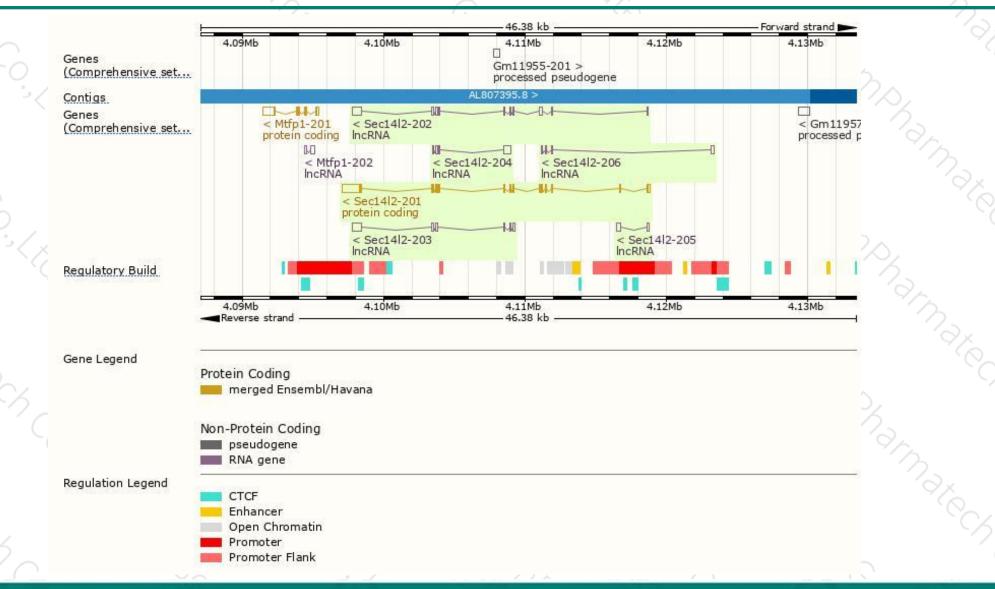
< Sec14l2-201 protein coding

Reverse strand

- 21.79 kb -

## **Genomic location distribution**





#### 江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.

## **Protein domain**

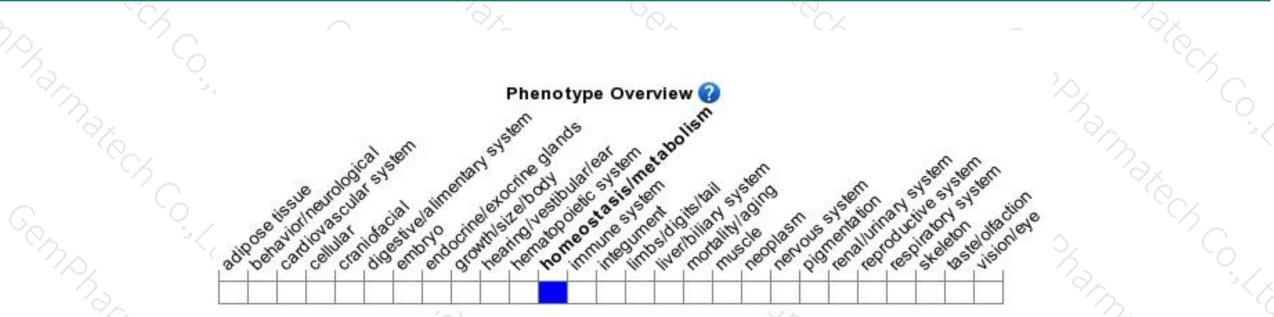
江苏



萃药康生物科技股份有限公	L	2834.		Cor	mPharmate	ch Co. Itd				Â	00-9660	0000
Scale bar	0	40	80	120	160	200	240	280	320	360	403	
		sense vari onymous v										
Variant Legend	2	7C).	Concerne and	U.							-	
All sequence SNPs/i	Sequence	e variants	(dbSNP and	d all other so	ources)	1		1.1				
CDD				IO lipid bindin								ò
	and the space of the test fails			The second s				and a fact that is real and that the				~
Gene3D	n <u>en la naciona en Estan</u> t		ng domain si	uperfamily				2.60.120.	580			
	PTHR2332										-	
PANTHER	PTHR2332-	4									-	0
PROSITE profiles	CRAL-TRIO lipid binding domain							GOLD don		9		
<u>Pfam</u>			CRAL-TR	IO lipid bindii	ng domain							
Prints		PR00180	r, the success contact and									
		CRAI/TRIC	CRAL-TRI	O lipid bindin domain	g domain							
SMART	CRAL/TRIC	D, N-termin	ial domain si									
Superfamily	CRAL-TRIO lipid binding domain superfamily							GOLD don	amily			
ENSMUSP0000003						12 4.5					-	

## 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 null allele exhibit decreased cholesterol synthesis and plasma levels under fasting conditions compared to wild-type mice.



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



