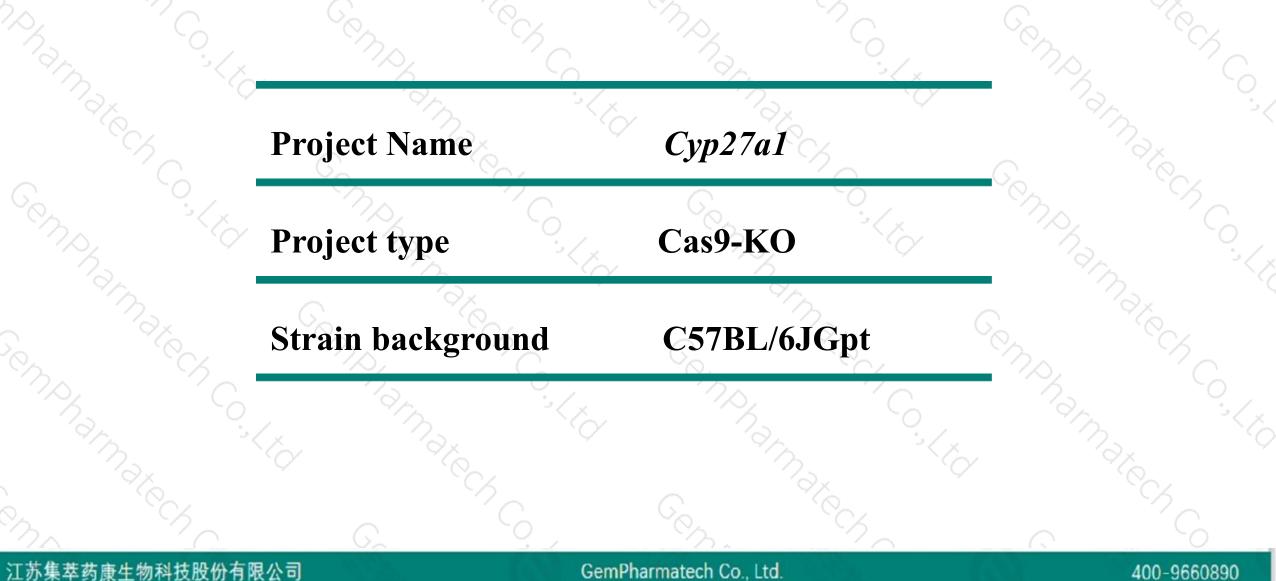


# Cyp27a1 Cas9-KO Strategy

Designer: Xueting Zhang Design Date: 2019-7-22

### **Project Overview**



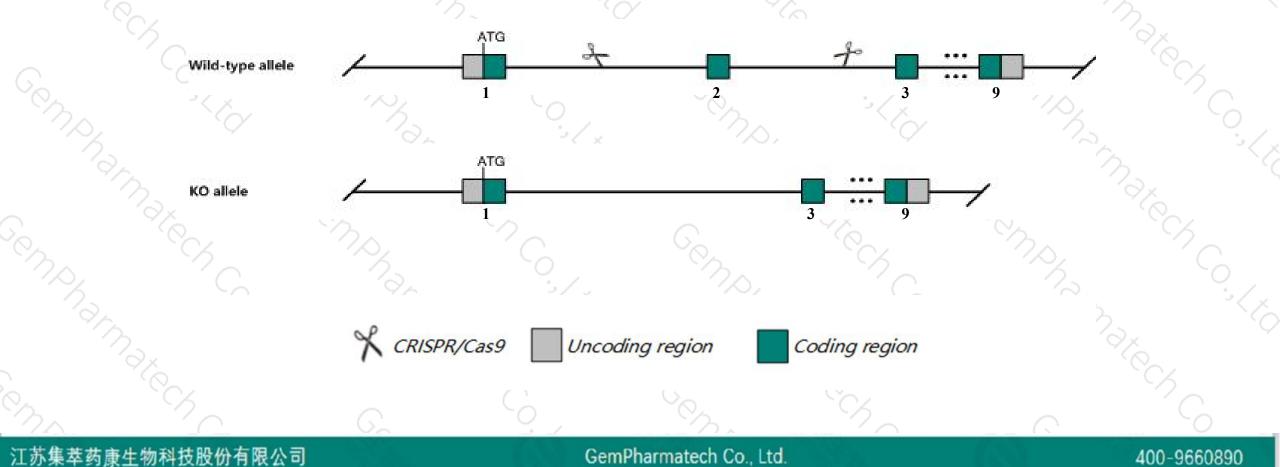


GemPharmatech Co., Ltd.

# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the Cyp27a1 gene. The schematic diagram is as follows:





- The Cyp27a1 gene has 3 transcripts. According to the structure of Cyp27a1 gene, exon2 of Cyp27a1-201 (ENSMUST0000027356.6) transcript is recommended as the knockout region. The region contains 191bp coding sequence. Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify Cyp27a1 gene. The brief process is as follows: CRISPR/Cas9 syst



- According to the existing MGI data, Homozygous null allele show hepato- and adrenomegaly, reduced bile acid synthesis, increased cholesterol 7alpha-hydroxylase activity and 7alpha-hydroxycholesterol levels, slightly higher 25-hydroxyvitamin D levels, and altered hepatic fatty acid, triacylglycerol, and adrenal cholesterol homeostasis.
- > The N-terminal of *Cyp27a1* gene will remain 87aa, it may remain the partial function of *Cyp27a1* gene.
- ➤ Transcript *Cyp27a1*-202&203 may not be affected.
- The *Cyp27a1* gene is located on the Chr1. 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.

#### 江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.

## **Gene information (NCBI)**



\$ ?

### Cyp27a1 cytochrome P450, family 27, subfamily a, polypeptide 1 [Mus musculus (house mouse)]

Gene ID: 104086, updated on 19-Mar-2019

#### Summary

Official Symbol	Cyp27a1 provided by MGI
	cytochrome P450, family 27, subfamily a, polypeptide 1 provided by MGI
Primary source	MGI:MGI:88594
See related	Ensembl:ENSMUSG0000026170
Gene type	protein coding
<b>RefSeq status</b>	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1300013A03Rik, Cyp27
Expression	Biased expression in liver adult (RPKM 72.0), bladder adult (RPKM 14.6) and 14 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.

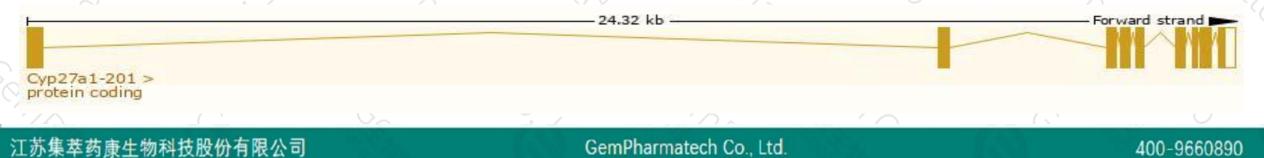
## **Transcript information (Ensembl)**



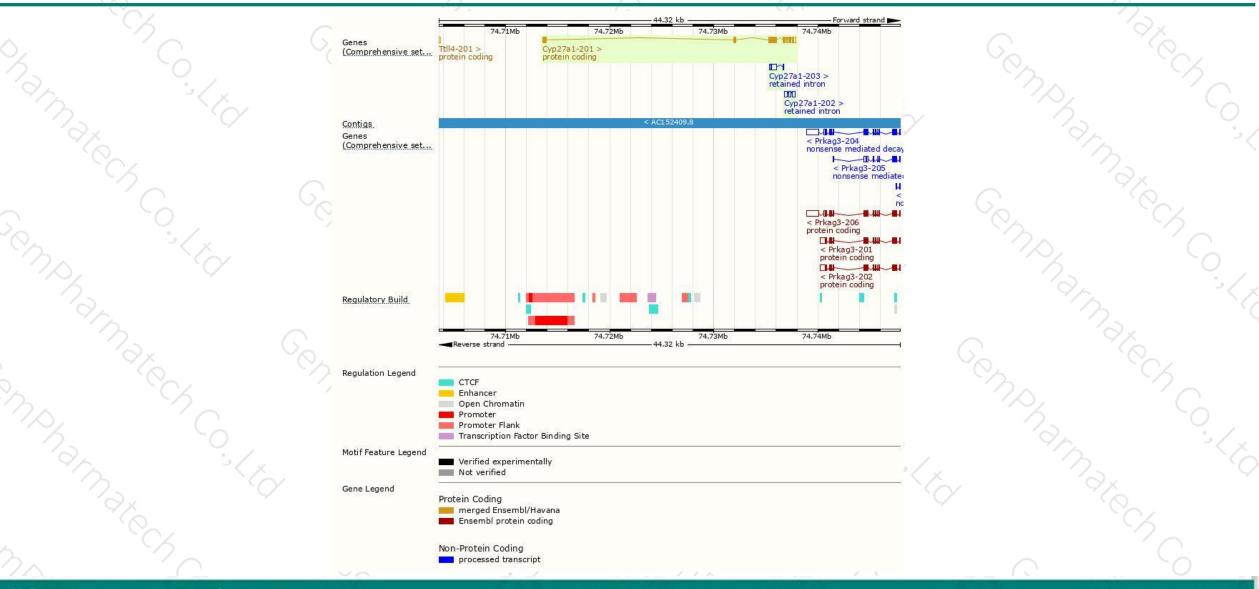
### The gene has 3 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags		
Cyp27a1-201	ENSMUST00000027356.6	1892	<u>533aa</u>	Protein coding	CCDS15054	Q9DBG1	TSL:1 GENCODE basic APPRIS P1		
Cyp27a1-202	ENSMUST00000189083.1	784	No protein	Retained intron	-		TSL:2		
Cyp27a1-203	ENSMUST00000190781.1	645	No protein	Retained intron	23	-	TSL:3		

The strategy is based on the design of *Cyp27a1-201* 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

集萃药康 GemPharmatech

### **Protein domain**



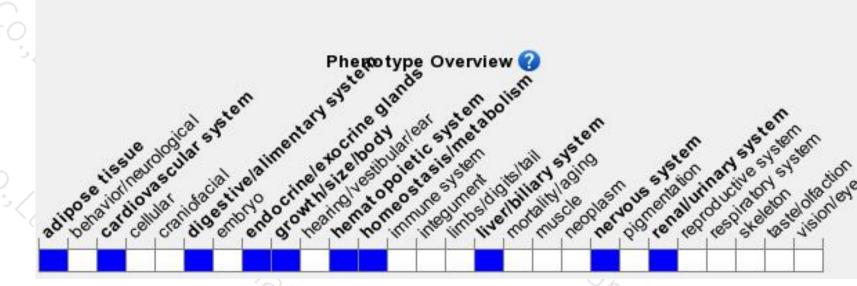
		<u>`C</u>		<u> </u>					
ENSMUSP00000027 MobiDB lite Low complexity (Seg)				17					
hmmpanther.	PTHR24291 :	3F6 ).							2
Superfamily domains	Cytochn	ome P450 superfa	mily						
Prints domain		Cytochrome P450	, E-class, gro	up I		ytochrome P45	0	_	2
Pfam domain	Cy	tochrome P450				former r			
PROSITE patterns	0.000							Cytochrom	e P45
Gene3D	Cytochro	me P450 superfar	nily						
All sequence SNPs/i	Sequence variants (	dbSNP and all o	ther sources	5)	0.0	(i) (i	.0	1.00	12
Variant Legend	missense varia								
Scale bar	<b>0</b> 60	120	180	240	300	360	420		533
~~~~~									

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

### 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,Homozygous null allele show hepato- and adrenomegaly, reduced bile acid synthesis, increased cholesterol 7alpha-hydroxylase activity and 7alpha-hydroxycholesterol levels, slightly higher 25-hydroxyvitamin D levels, and altered hepatic fatty acid, triacylglycerol, and adrenal cholesterol homeostasis.

#### 江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.



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



