Cyp8b1 Cas9-KO Strategy matech Co. 1 to Rondhamater Co-ty

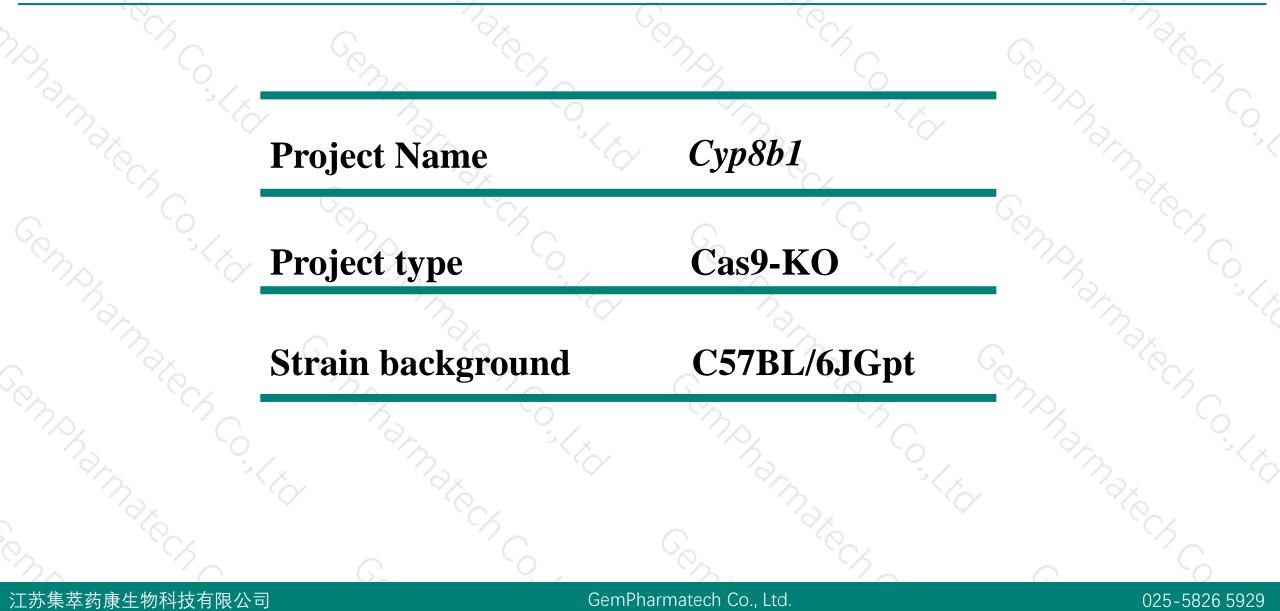
Designer: Enphamater Co. 1 to

Daohua Xu

Genphaman

Project Overview

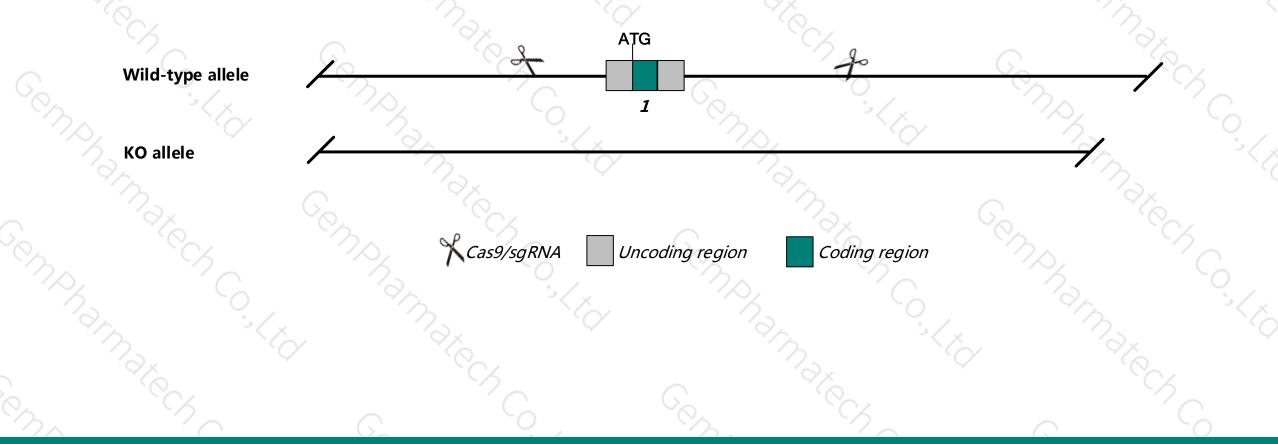




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cyp8b1* gene. The schematic diagram is as follows:



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- The Cyp8b1 gene has 1 transcript.According to the structure of Cyp8b1 gene, exon1 of Cyp8b1-201 (ENSMUST0000062474.4) transcript is recommended as the knockout region.The region contains all coding sequence.Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cyp8b1* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice



- According to the existing MGI data, Mice homozygous for a knock-out allele lack synthsesis of cholate (a primary bile acid) and its metabolites and display an increased bile acid pool and alterations in cholesterol metabolism.
- The Cyp8b1 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, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



2 ?

Cyp8b1 cytochrome P450, family 8, subfamily b, polypeptide 1 [Mus musculus (house mouse)]

Gene ID: 13124, updated on 23-Feb-2019

Summary

Official SymbolCyp8b1 provided by MGIOfficial Full Namecytochrome P450, family 8, subfamily b, polypeptide 1 provided by MGIPrimary sourceMGI:MGI:1338044See relatedEnsembl:ENSMUSG0000050445Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Muroidea; Murinae; Mus; MusOrthologshuman all

Transcript information (Ensembl)



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

Show/hide columns (1 hidden)							Filter		
Name 🍦	Transcript ID 💧	bp 🍦	Protein 🖕	Biotype	CCDS 🖕	UniProt 🍦	Flags 🖕		
Cyp8b1-201	ENSMUST0000062474.4	1950	<u>500aa</u>	Protein coding	<u>CCDS23641</u> &	<u>A0A0R4J0N7</u> &	TSL:NA	GENCODE basic	APPRIS P1

The strategy is based on the design of Cyp8b1-201 transcript, The transcription is shown below

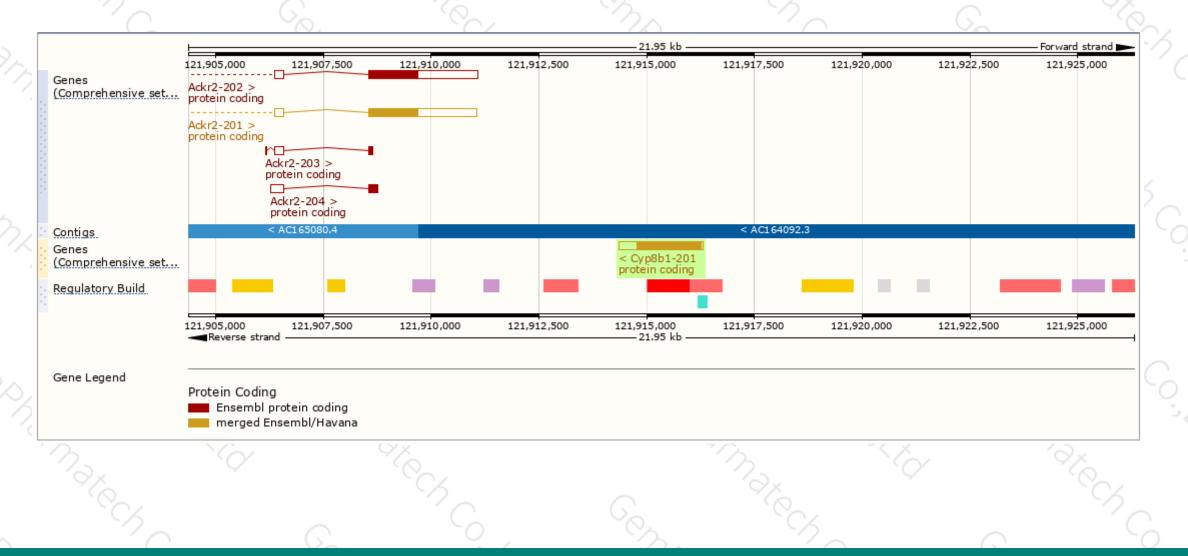
1			/ A 1		
	< Cyp8b1-201 protein coding Reverse strand		1.95 kb		
	Reverse strand	5 1	1.95 KD		

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Genomic location distribution





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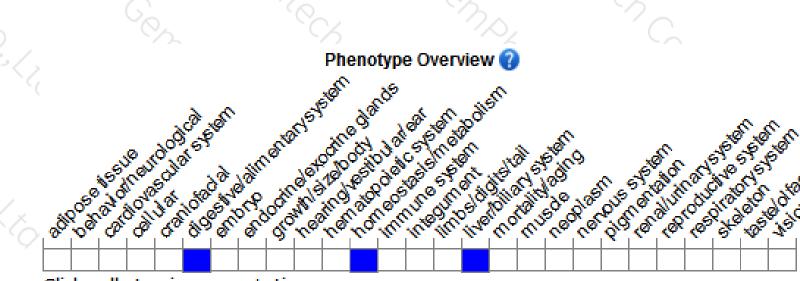
Protein domain



0	$\sim \gamma_{\rm S}$	^S C	No.	C.		<u>`</u> %
ENSMUSP00000052 Transmembrane heli Low complexity (Seg)			_			_
hmmpanther	Cytochrome P450 8B1 PTHR24306					
Superfamily domains	Cytochrome P450 superfamily					
Prints domain Pfam. domain	Cytochrome P450, E-class, grou Cytochrome P450	ıp IV				2
PIRSF domain	Cytochrome P450, cholesterol 7-alpha-mon	ooxygenase-type				
Gene3D	Cytochrome P450 8B1 Cytochrome P450 superfamily					
All sequence SNPs/i	Sequence variants (dbSNP and all othe	r sources)	111	11 11	1 1 1 1	1
Variant Legend	missense variant		synon			
Scale bar	o 60	120 180	240	300 3	60 420	500
		~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~	Nor.	$\sim^{2}$ C		, Ъ.

# Mouse phenotype description(MGI)





Click cells to view annotations.

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 knock-out allele lack synthsesis of cholate (a primary bile acid) and its metabolites and display an increased bile acid pool and alterations in cholesterol metabolism.

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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



