

Apoh Cas9-CKO Strategy

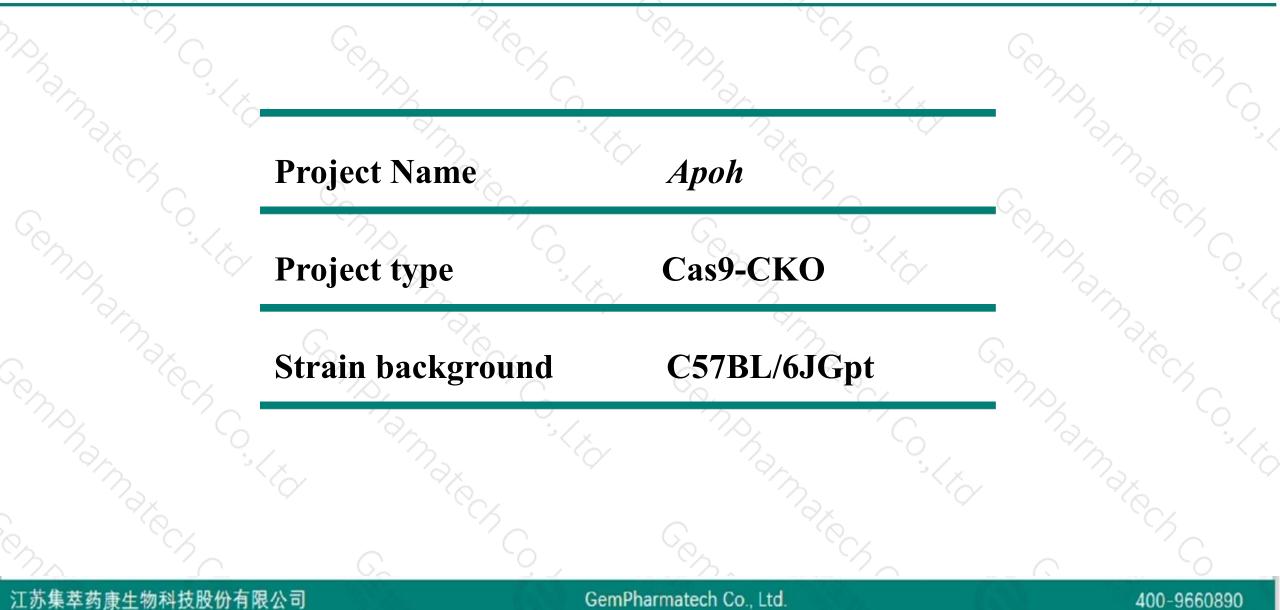
Designer: Reviewer: Design Date:

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Yang Zeng Jia Yu 2020-2-10

Project Overview



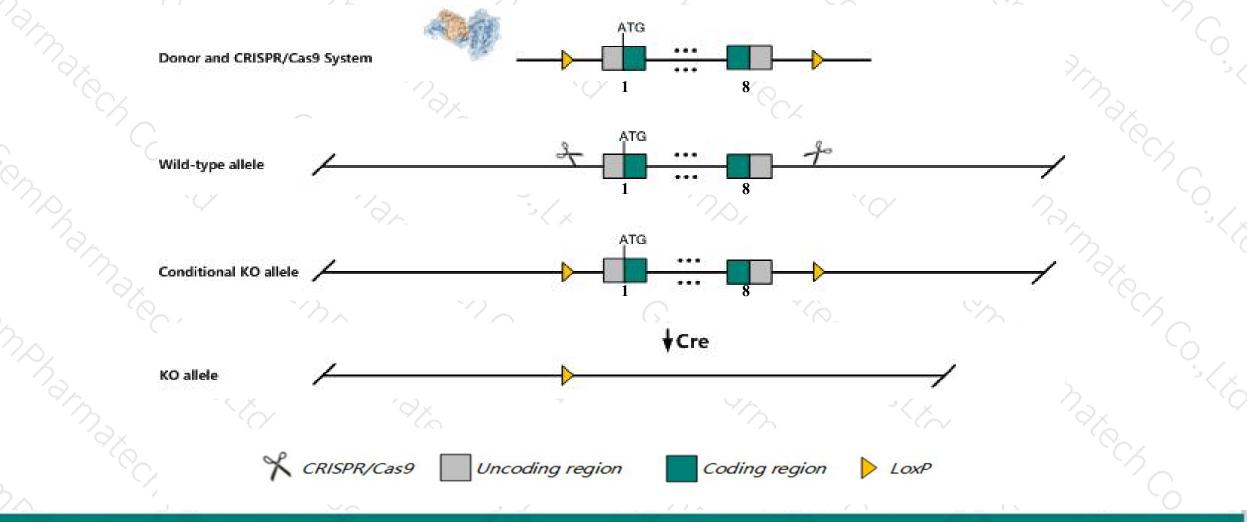


Conditional Knockout strategy



400-9660890

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



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The Apoh gene has 4 transcripts. According to the structure of Apoh gene, exon1-exon8 of Apoh-201 (ENSMUST0000000049.5) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.

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



- According to the existing MGI data, Homozygous mutation of this gene results in reduced viability and reduced thrombin production. Only 8% homozygous null animals are born from heterozygous intercrosses.
- The Apoh 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.

Gene information (NCBI)



☆ ?

Apoh apolipoprotein H [Mus musculus (house mouse)]

Gene ID: 11818, updated on 31-Jan-2019

Summary

Official SymbolApoh provided by MGIOfficial Full Nameapolipoprotein H provided by MGIPrimary sourceMGI:MGI:88058See relatedEnsembl:ENSMUSG0000000049Gene typeprotein codingprotein codingVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Muroidea; Murinae; Mus; MusAlso knownasB2GPI, beta-2-GPI, beta2-GPIExpressionBiased expression in liver E18 (RPKM 1780.2), liver adult (RPKM 813.8) and 2 other tissues
See more

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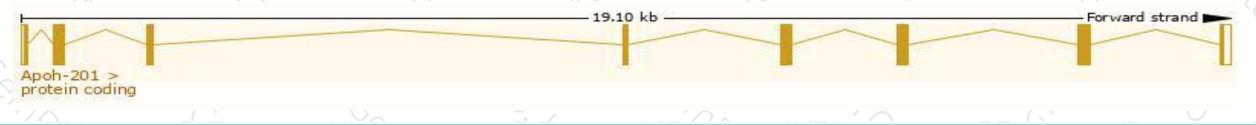
Transcript information (Ensembl)



The gene has 4 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Apoh-201	ENSMUST0000000049.5	1190	<u>345aa</u>	Protein coding	CCDS25574	<u>Q01339</u>	TSL:1 GENCODE basic APPRIS P1
Apoh-204	ENSMUST00000152958.7	731	<u>154aa</u>	Protein coding		<u>17HJR3</u>	CDS 3' incomplete TSL:5
Apoh-203	ENSMUST00000146050.1	374	<u>33aa</u>	Protein coding	20	<u>J3JS85</u>	CDS 3' incomplete TSL:2
Apoh-202	ENSMUST00000133383.7	367	<u>51aa</u>	Protein coding	2	17HPW5	CDS 3' incomplete TSL:3

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

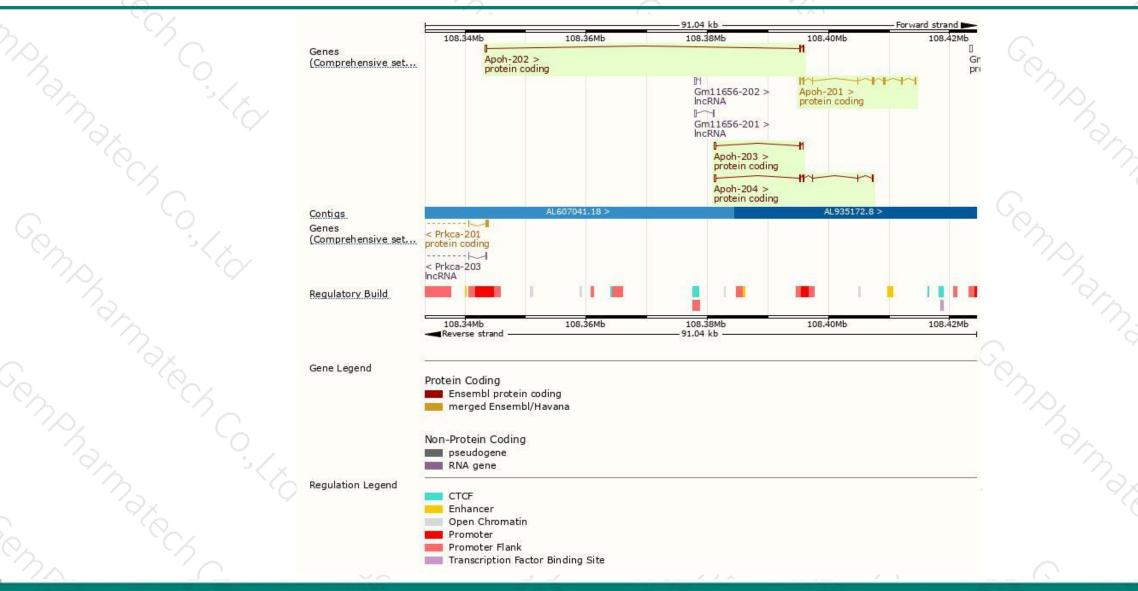


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Genomic location distribution





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Protein domain



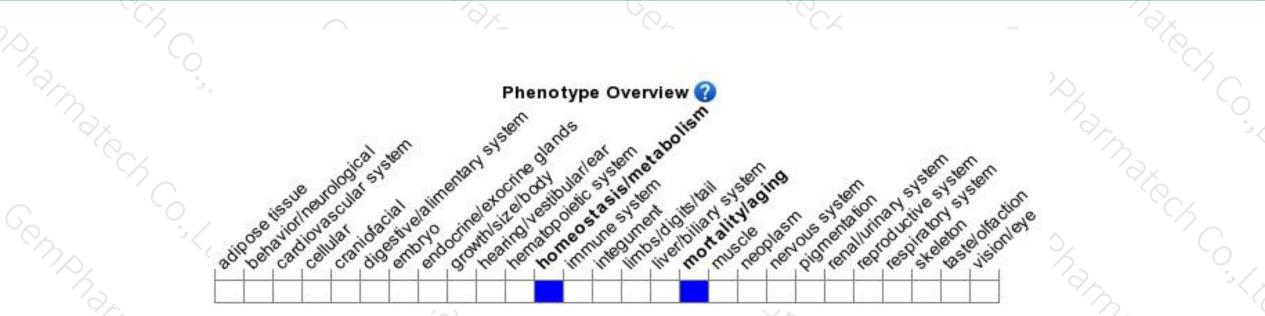
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Pfam	Sushi/SCR/CC	P domain					Beta-2	2-glycoprotei	in-1 fifth domai
PROSITE profiles	Sushi/SCR/CCF	domain			100			100	9
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	PTHR19325								
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Mouse 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 mutation of this gene results in reduced viability and reduced thrombin production. Only 8% homozygous null animals are born from heterozygous intercrosses.



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



