

Coro2b Cas9-KO Strategy

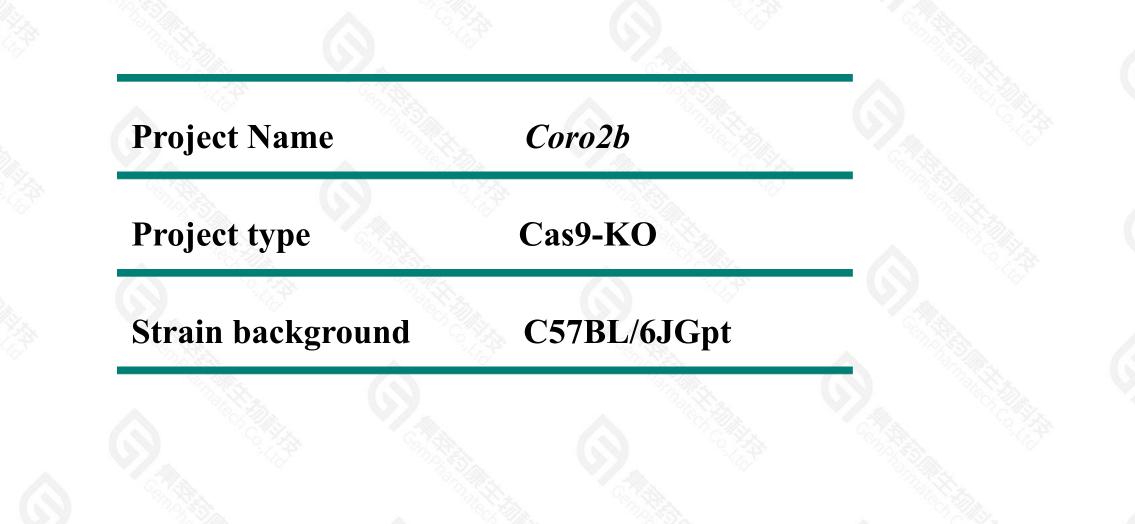
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Reviewer: Lingyan Wu

Design Date: 2021-7-9

Project Overview





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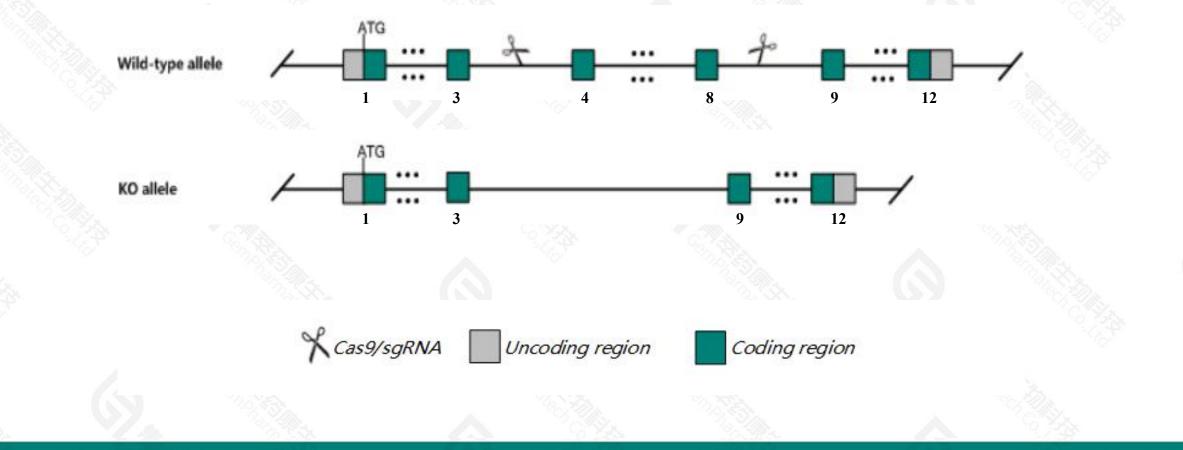
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Knockout strategy



400-9660890

This model will use CRISPR/Cas9 technology to edit the *Coro2b* gene. The schematic diagram is as follows:



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> The *Coro2b* gene has 7 transcripts. According to the structure of *Coro2b* gene, exon4-exon8 of *Coro2b*-201(ENSMUST00000048043.12) transcript is recommended as the knockout region. The region contains 634bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Coro2b* 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.



- According to the existing MGI data, homozygous knockout reduces susceptibility to Doxorubicin-induced focal segmental glomerulosclerosis with lower levels of proteinuria and less renal glomerulus damage.
- > The *Coro2b* 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,all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

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Gene information (NCBI)

Coro2b coronin, actin binding protein, 2B [Mus musculus (house mouse)]

Gene ID: 235431, updated on 17-Dec-2020

Summary

Official Symbol	Coro2b provided by MGI
Official Full Name	coronin, actin binding protein, 2B provided by MGI
Primary source	MGI:MGI:2444283
See related	Ensembl:ENSMUSG0000041729
Gene type	protein coding
RefSeq status	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	CLIPINC, E130012P22Rik
Expression	Broad expression in cerebellum adult (RPKM 45.9), cortex adult (RPKM 26.6) and 16 other tissuesSee more
Orthologs	human all



☆ ?

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Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

Name Transcript ID		Protein	Biotype	CCDS	UniProt	Flags		
ENSMUST0000048043.12	3610	<u>480aa</u>	Protein coding	CCDS23264		TSL:1 , GENCODE basic , APPRIS P1 ,		
ENSMUST00000164246.9	987	<u>317aa</u>	Protein coding	-		CDS 3' incomplete , TSL:5 ,		
ENSMUST00000131981.2	618	<u>181aa</u>	Protein coding	10		CDS 5' incomplete , TSL:3 ,		
ENSMUST00000173171.3	396	<u>121aa</u>	Protein coding	17		CDS 3' incomplete , TSL:5 ,		
ENSMUST00000174439.2	393	<u>94aa</u>	Protein coding	14		CDS 3' incomplete , TSL:5 ,		
ENSMUST00000123379.2	1872	No protein	Retained intron	(ī)		TSL:1,		
ENSMUST00000151604.2	816	No protein	Retained intron	-		TSL:3,		
	ENSMUST0000048043.12 ENSMUST00000164246.9 ENSMUST00000131981.2 ENSMUST00000173171.3 ENSMUST00000174439.2	ENSMUST0000048043.12 3610 ENSMUST00000164246.9 987 ENSMUST00000131981.2 618 ENSMUST00000173171.3 396 ENSMUST00000174439.2 393 ENSMUST00000123379.2 1872	ENSMUST0000048043.12 3610 480aa ENSMUST0000164246.9 987 317aa ENSMUST0000131981.2 618 181aa ENSMUST0000173171.3 396 121aa ENSMUST0000174439.2 393 94aa ENSMUST0000123379.2 1872 No protein	ENSMUST0000048043.123610480aaProtein codingENSMUST00000164246.9987317aaProtein codingENSMUST00000131981.2618181aaProtein codingENSMUST00000173171.3396121aaProtein codingENSMUST00000174439.239394aaProtein codingENSMUST00000123379.21872No proteinRetained intron	ENSMUST0000048043.123610480aaProtein codingCCDS23264ENSMUST0000164246.9987317aaProtein coding-ENSMUST0000131981.2618181aaProtein coding-ENSMUST0000173171.3396121aaProtein coding-ENSMUST0000174439.239394aaProtein coding-ENSMUST0000123379.21872No proteinRetained intron-	ENSMUST0000048043.123610480aaProtein codingCCDS23264ENSMUST0000164246.9987317aaProtein coding-ENSMUST0000131981.2618181aaProtein coding-ENSMUST0000173171.3396121aaProtein coding-ENSMUST0000174439.239394aaProtein coding-ENSMUST0000123379.21872No proteinRetained intron-		

The strategy is based on the design of Coro2b-201 transcript, the transcription is shown below:

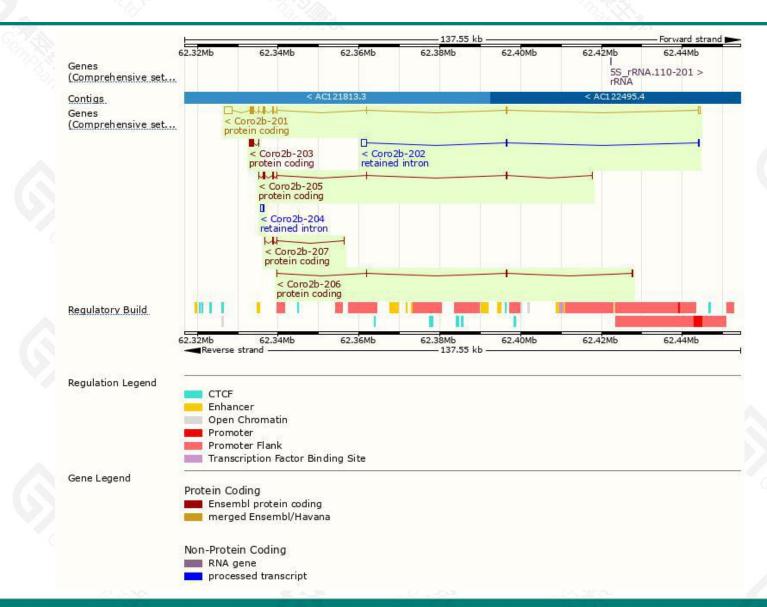


- 117.55 kb -

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Genomic location distribution





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Protein domain

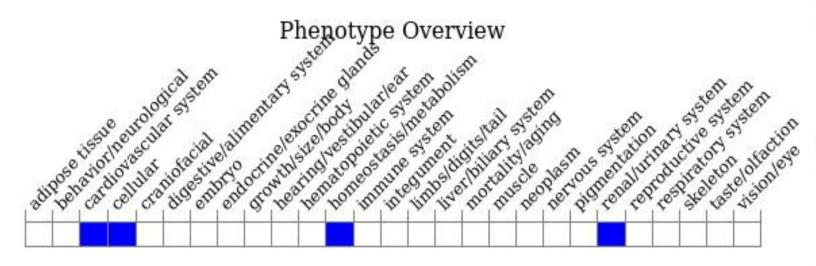


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Mouse phenotype description(MGI)



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 knockout reduces susceptibility to Doxorubicin-induced focal segmental glomerulosclerosis with lower levels of proteinuria and less renal glomerulus damage.



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



