

Sema3d Cas9-CKO Strategy

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Reviewer:

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Design Date:

2019-9-19

Project Overview



Project Name

Sema3d

Project type

Cas9-CKO

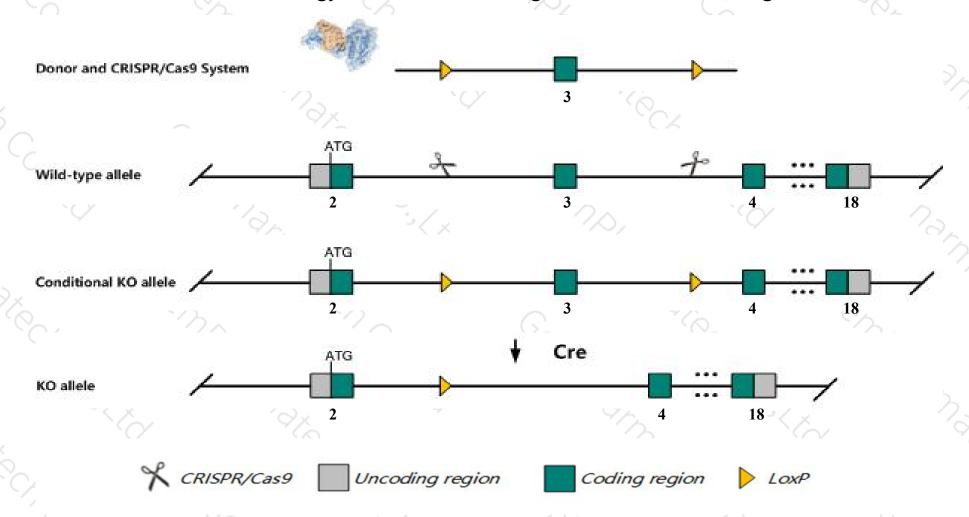
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Sema3d gene. The schematic diagram is as follows:



Technical routes



- The Sema3d gene has 5 transcripts. According to the structure of Sema3d gene, exon3 of Sema3d-201 (ENSMUST00000030868.10) transcript is recommended as the knockout region. The region contains 161bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Sema3d* 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 knock-out allele exhibit pulmonary vein connection to the right atrium and atrial septal defect.
- The *Sema3d* gene is located on the Chr5. 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)



Sema3d sema domain, immunoglobulin domain (Ig), short basic domain, secreted, (semaphorin) 3D [
Mus musculus (house mouse)]

Gene ID: 108151, updated on 12-Aug-2019

Summary



Official Symbol Sema3d provided by MGI

Official Full Name sema domain, immunoglobulin domain (Ig), short basic domain, secreted, (semaphorin) 3D provided by MGI

Primary source MGI:MGI:1860118

See related Ensembl: ENSMUSG00000040254

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 4631426B19Rik

Expression Broad expression in limb E14.5 (RPKM 4.3), bladder adult (RPKM 3.0) and 18 other tissues See more

Orthologs <u>human</u> all

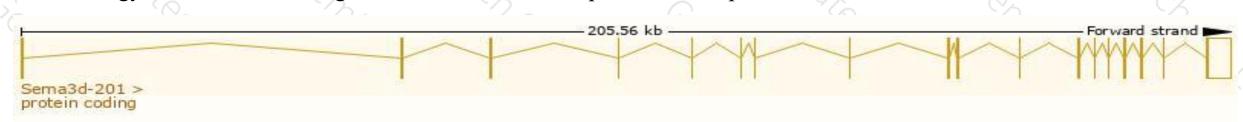
Transcript information (Ensembl)



The gene has 5 transcripts, all transcripts are shown below:

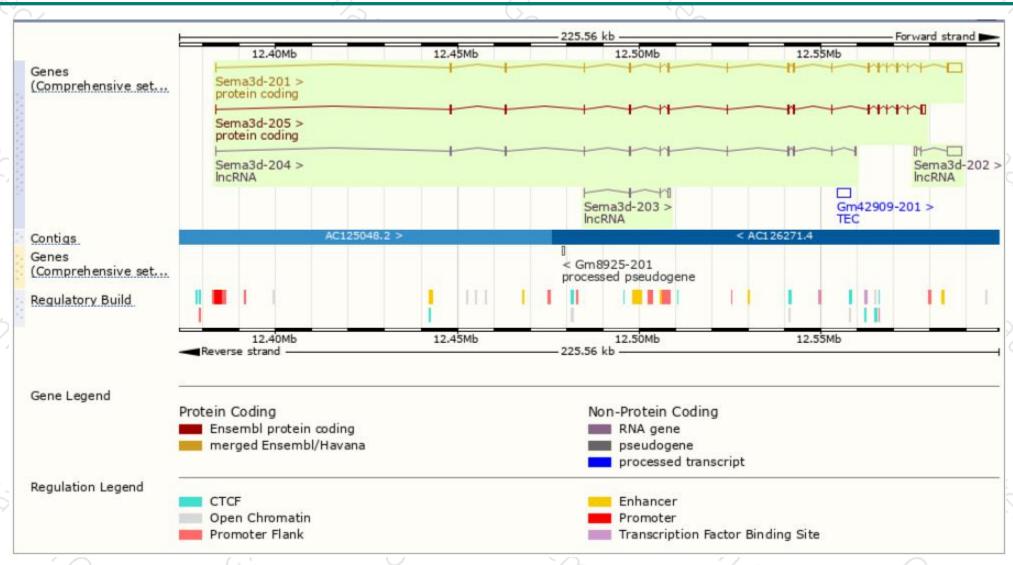
Name 🍦	Transcript ID	bp 🛊	Protein 🍦	Biotype	CCDS 🍦	UniProt	Flags
Sema3d-201	ENSMUST00000030868.10	6307	<u>777aa</u>	Protein coding	CCDS39013 ₽	Q8BH34 €	TSL:1 GENCODE basic APPRIS P1
Sema3d-205	ENSMUST00000197927.1	3207	<u>660aa</u>	Protein coding	12	A0A0G2JDP8₺	TSL:1 GENCODE basic
Sema3d-202	ENSMUST00000195923.1	4498	No protein	IncRNA	2	12	TSL:1
Sema3d-204	ENSMUST00000196618.4	1808	No protein	IncRNA		-	TSL:1
Sema3d-203	ENSMUST00000196093.1	708	No protein	IncRNA	15	-	TSL:5

The strategy is based on the design of Sema3d-201 transcript, The transcription is shown below



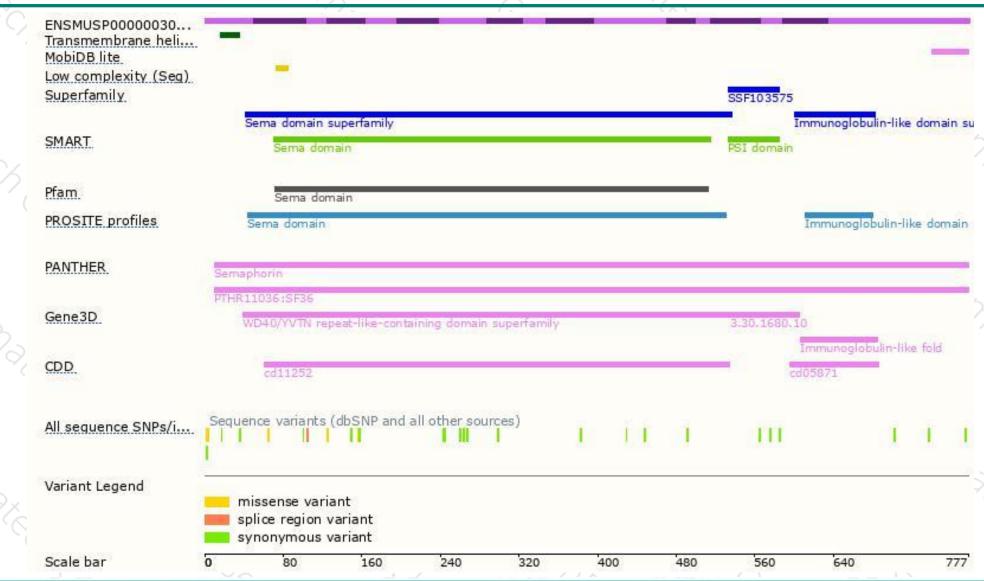
Genomic location distribution





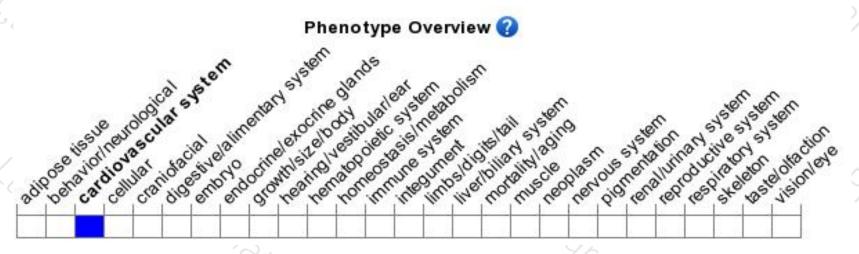
Protein domain





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 knock-out allele exhibit pulmonary vein connection to the right atrium and atrial septal defect.



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





