Sema5a Cas9-CKO Strategy

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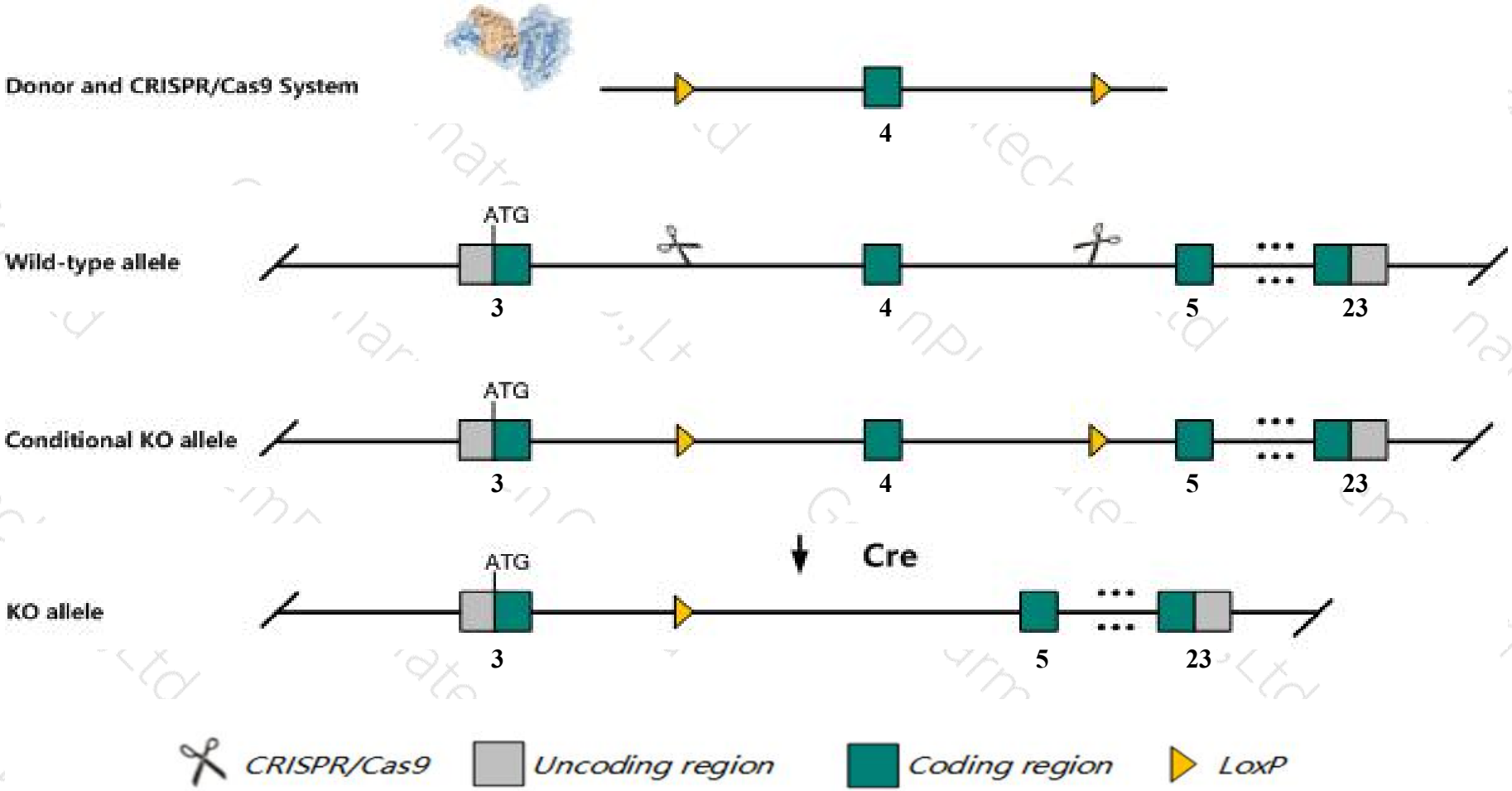
Project Overview

Project Name	***Sema5a***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sema5a* gene. The schematic diagram is as follows:



Technical routes

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- The *Sema5a* gene has 9 transcripts. According to the structure of *Sema5a* gene, exon4 of *Sema5a-201* (ENSMUST00000067458.6) transcript is recommended as the knockout region.The region contains 100bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Sema5a* 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 one null mutation die during organogenesis and display defects in branching of cranial vessels. Mice homozygous for another null mutation appear normal.

- The *Sema5a* gene is located on the Chr15. 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）

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Sema5a sema domain, seven thrombospondin repeats (type 1 and type 1-like), transmembrane domain (TM) and short cytoplasmic domain, (semaphorin) 5A [Mus musculus (house mouse)]

Gene ID: 20356, updated on 12-Mar-2019

Summary

Official Symbol	Sema5a provided by MGI
Official Full Name	sema domain, seven thrombospondin repeats (type 1 and type 1-like), transmembrane domain (TM) and short cytoplasmic domain, (semaphorin) 5A provided by MGI
Primary source	MGI:MGI:107556
See related	Ensembl:ENSMUSG00000022231
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	5930434A13, 9130201M22Rik, AI464145, Semaf, semF
Expression	Broad expression in ovary adult (RPKM 13.8), limb E14.5 (RPKM 11.9) and 21 other tissues See more
Orthologs	human all

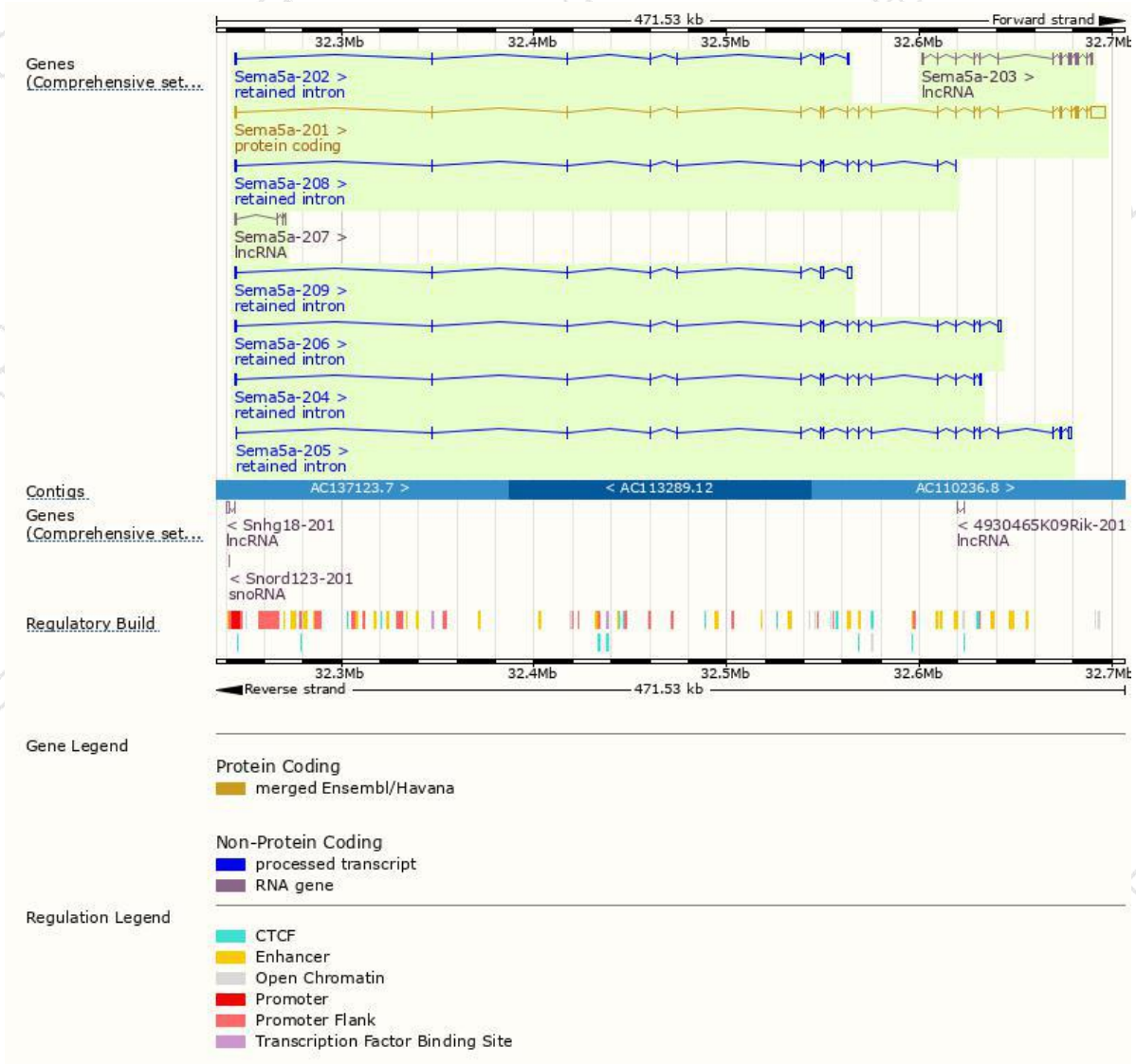
Transcript information（Ensembl）

The gene has 9 transcripts,all transcripts are shown below:

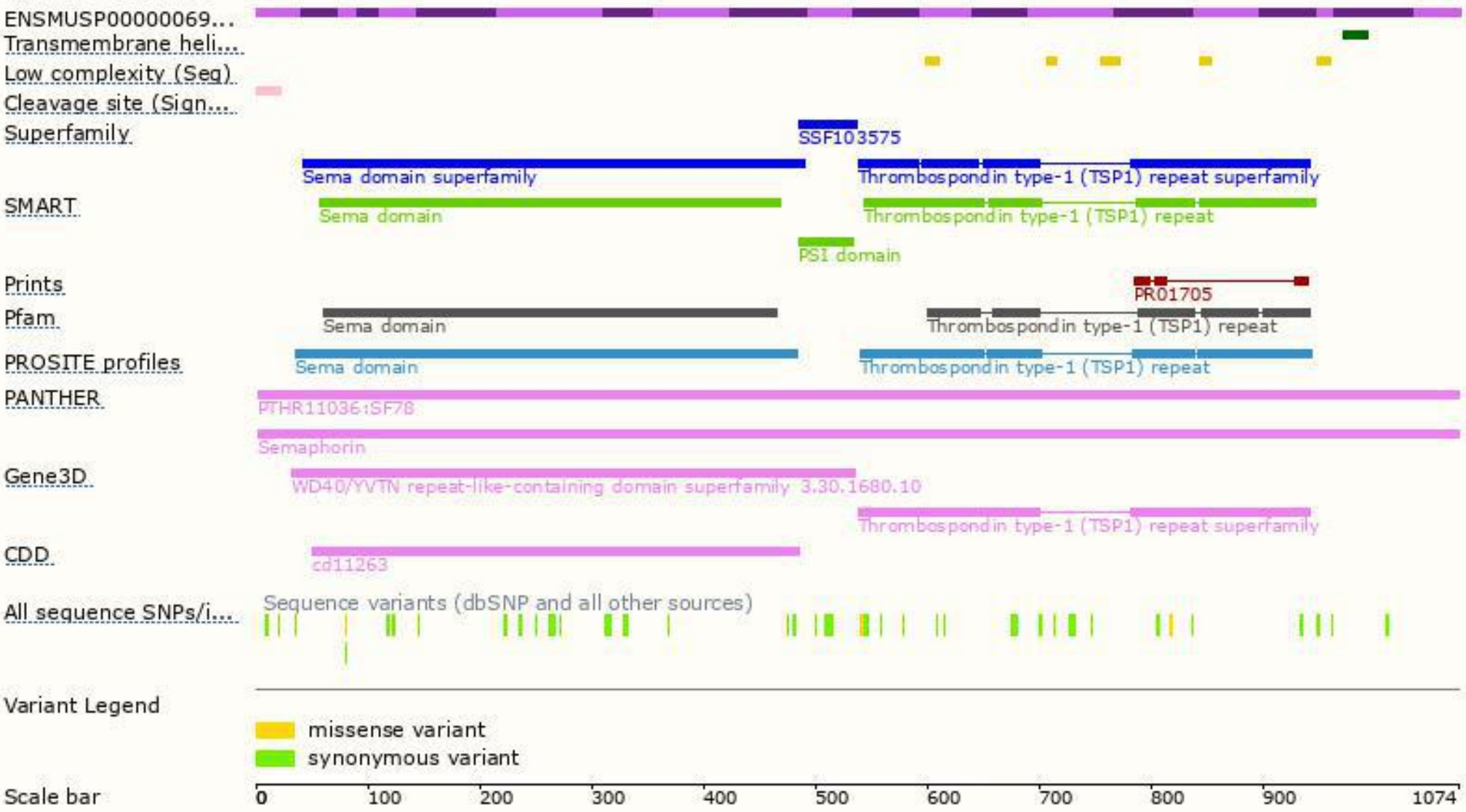
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sema5a-201	ENSMUST00000067458.6	10809	1074aa	Protein coding	CCDS37055	Q3UPZ0	TSL:1 GENCODE basic APPRIS P1
Sema5a-209	ENSMUST00000228555.1	5442	No protein	Retained intron	-	-	
Sema5a-206	ENSMUST00000228015.1	3951	No protein	Retained intron	-	-	
Sema5a-205	ENSMUST00000227976.1	3725	No protein	Retained intron	-	-	
Sema5a-204	ENSMUST00000227802.1	3158	No protein	Retained intron	-	-	
Sema5a-208	ENSMUST00000228442.1	2571	No protein	Retained intron	-	-	
Sema5a-202	ENSMUST00000226876.1	2375	No protein	Retained intron	-	-	
Sema5a-203	ENSMUST00000227429.1	2774	No protein	lncRNA	-	-	
Sema5a-207	ENSMUST00000228103.1	902	No protein	lncRNA	-	-	

The strategy is based on the design of *Sema5a-201* transcript,The transcription is shown below

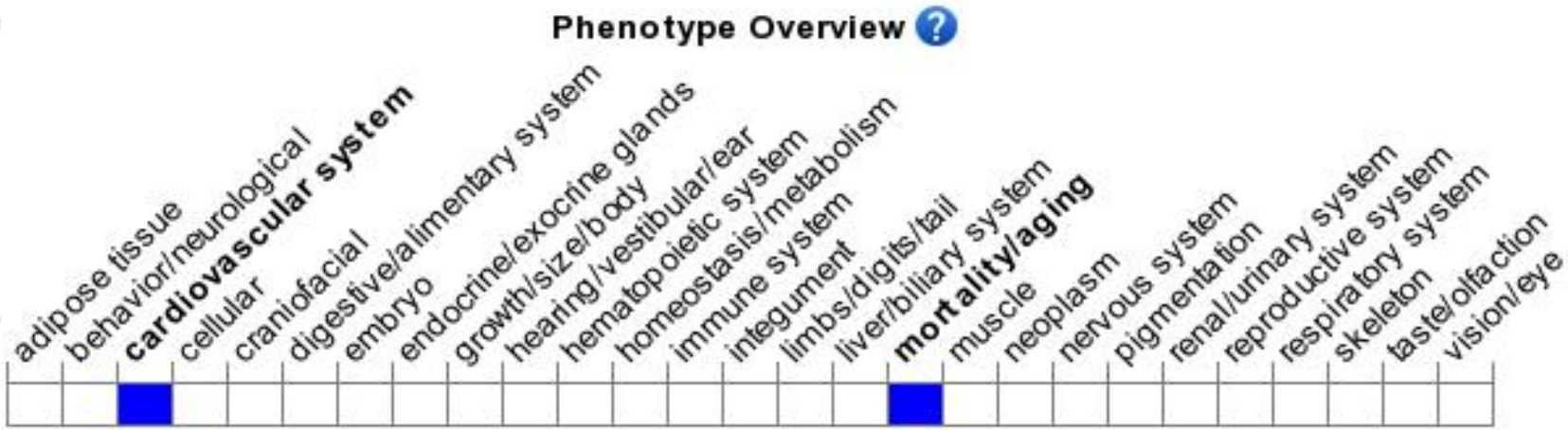
Genomic location distribution



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/).

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If you have any questions, you are welcome to inquire.

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