Sema4c Cas9-CKO *Strategy*

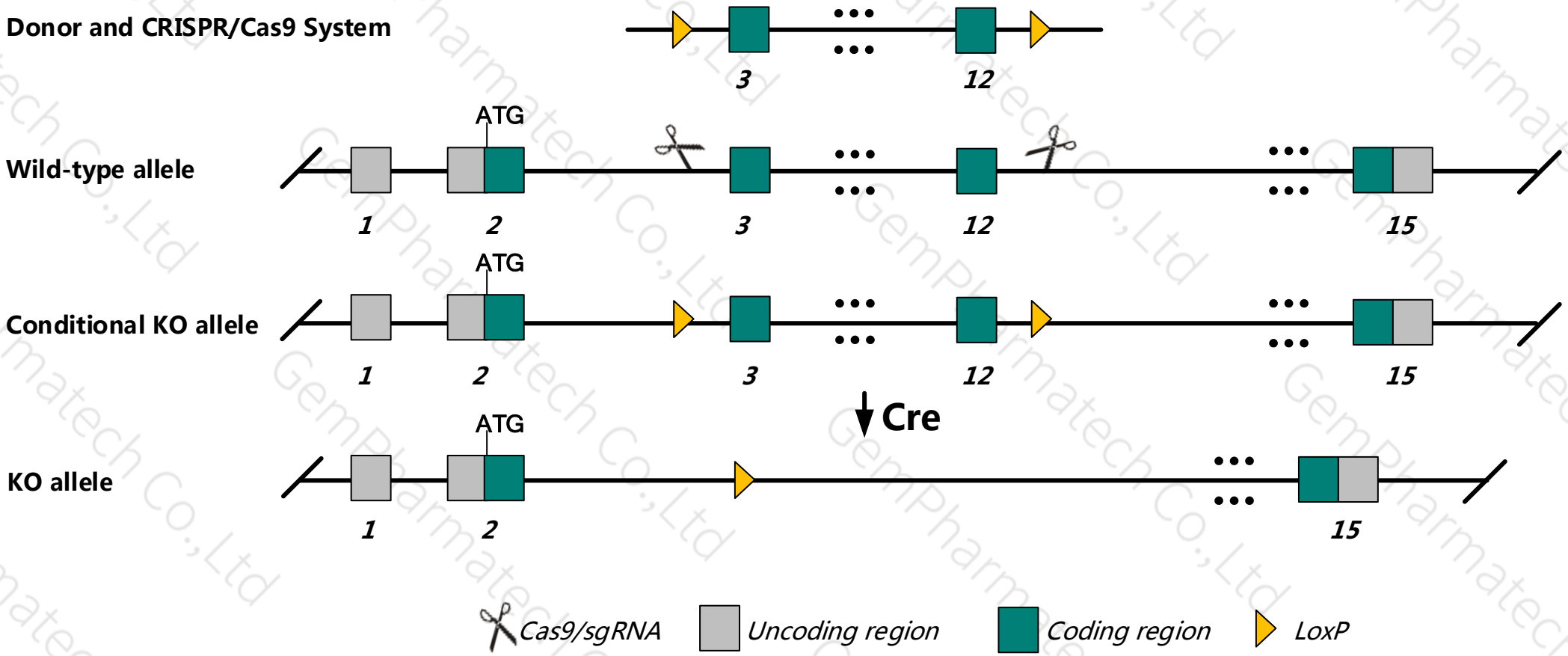
Designer: **Daohua Xu**
Reviewer： **Huimin Su**
Design Date: **2019-9-28**

Project Overview

Project Name	*Sema4c*
Project type	Cas9-CKO
Animal background	C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sema4c* gene. The schematic diagram is as follows:



Technical routes

- The *Sema4c* gene has 8 transcripts, According to the structure of *Sema4c* gene, exon3-exon12 of *Sema4c-201* transcript is recommended as the knockout region.The region contains the 1334bp coding seqence.Knock out the region,result in destruction of protein.

- This project uses CRISPR/Cas9 technology to modify *Sema4c* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed, Cas9, sgRNA and donor were microinjected into fertilized eggs of C57BL/6JGpt mice and homologous recombination was carried out to obtain F0 mice. A stable and hereditary F1 generation mouse model was obtained by mating F0 generation mice with C57BL/6JGpt mice which were confirmed positive by PCR-sequencing.

- The flox mice was 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 targeted mutation exhibit exencephaly, neonatal lethality, and abnormal cerebellum morphology.

- The *Sema4c* gene is located in the Chr1. If the knockout mice are mixed with other mice, two target genes are avoided on the same chromosome as possible, otherwise the offspring of mice with double gene positive and homozygous gene knockout can not be obtained.

- This Strategy is designed based on genetic information in existing databases. Due to the complexity of gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information （NCBI）

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Sema4c sema domain, immunoglobulin domain (Ig), transmembrane domain (TM) and short cytoplasmic domain, (semaphorin) 4C [*Mus musculus* (house mouse)]

Gene ID: 20353, updated on 1-Jan-2019

Summary

Official Symbol	Sema4c provided by MGI
Official Full Name	sema domain, immunoglobulin domain (Ig), transmembrane domain (TM) and short cytoplasmic domain, (semaphorin) 4C provided by MGI
Primary source	MGI:MGI:109252
See related	Ensembl:ENSMUSG00000026121
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Semaf; Semai; sema I; Semacl1; AI426163; M-Sema F
Summary	This gene encodes a member of the semaphorin family of proteins that have diverse functions in neuronal development, heart morphogenesis, vascular growth, tumor progression and immune cell regulation. Lack of the encoded protein in some mice causes exencephaly resulting in neonatal lethality. Mice that bypass exencephaly show no obvious behavioral defects but display distinct pigmentation defects. Alternative splicing of this gene results in multiple transcript variants. [provided by RefSeq, Jan 2015]
Expression	Ubiquitous expression in CNS E11.5 (RPKM 14.0), CNS E14 (RPKM 13.4) and 27 other tissues See more
Orthologs	human all

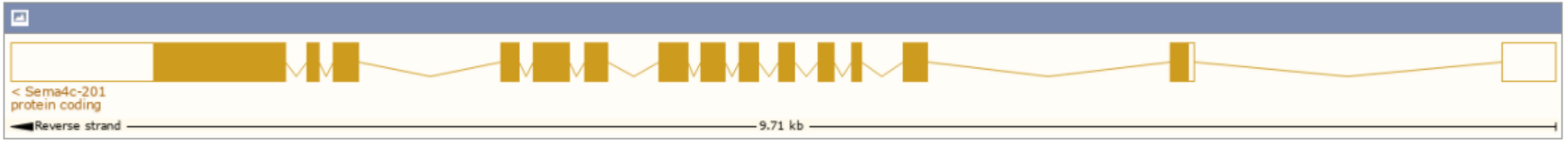
Transcript information （Ensembl）

The gene has 8 transcripts, and all transcripts are shown below ：

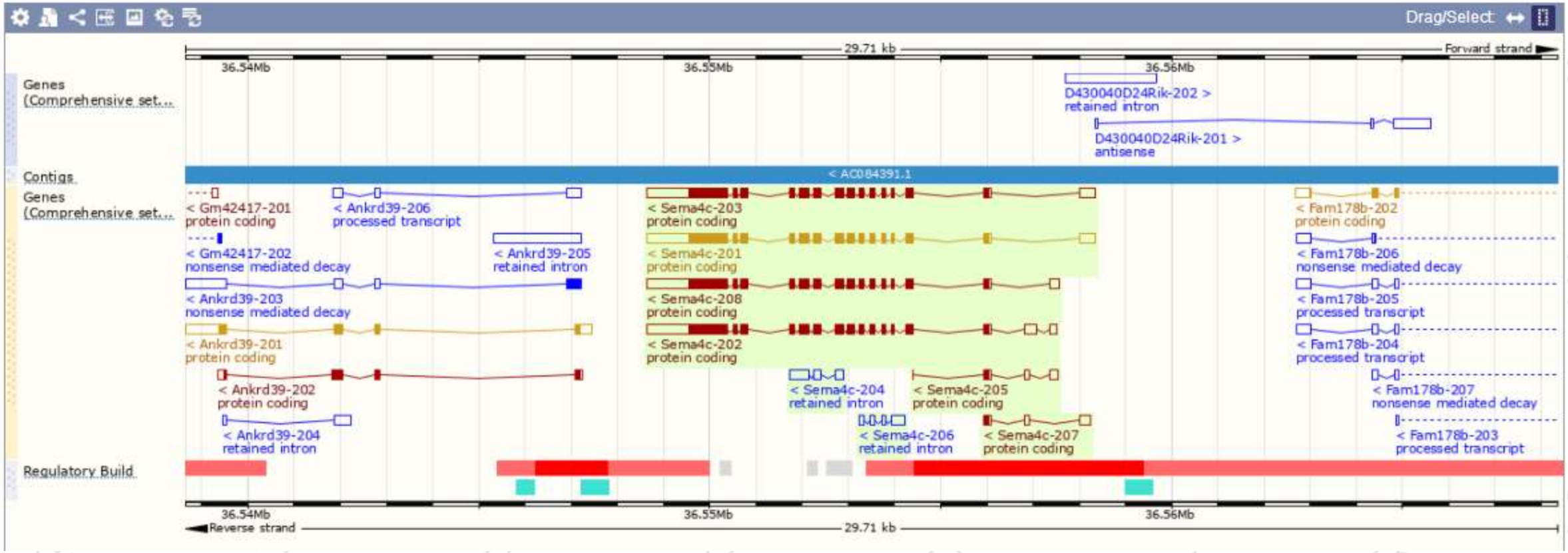
Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Sema4c-202	ENSMUST00000191642.5	3892	834aa	Protein coding	CCDS48240	Q64151	NM_001304330 NP_001291259	TSL:5 GENCODE basic APPRIS P1
Sema4c-203	ENSMUST00000191677.5	3779	834aa	Protein coding	CCDS48240	Q64151	-	TSL:5 GENCODE basic APPRIS P1
Sema4c-201	ENSMUST00000114991.7	3773	834aa	Protein coding	CCDS48240	Q64151	NM_001126047 NP_001119519	TSL:1 GENCODE basic APPRIS P1
Sema4c-208	ENSMUST00000195620.5	3640	834aa	Protein coding	CCDS48240	Q64151	NM_001304329 NP_001291258	TSL:1 GENCODE basic APPRIS P1
Sema4c-207	ENSMUST00000195339.2	508	36aa	Protein coding	-	A0A0A6YX48	-	CDS 3' incomplete TSL:1
Sema4c-205	ENSMUST00000193382.5	452	37aa	Protein coding	-	A0A0A6YXK9	-	CDS 3' incomplete TSL:5
Sema4c-204	ENSMUST00000191785.1	745	No protein	Retained intron	-	-	-	TSL:3
Sema4c-206	ENSMUST00000195160.1	618	No protein	Retained intron	-	-	-	TSL:3

The strategy is based on the design of *Sema4c-201* transcript,The transcription is shown below ：

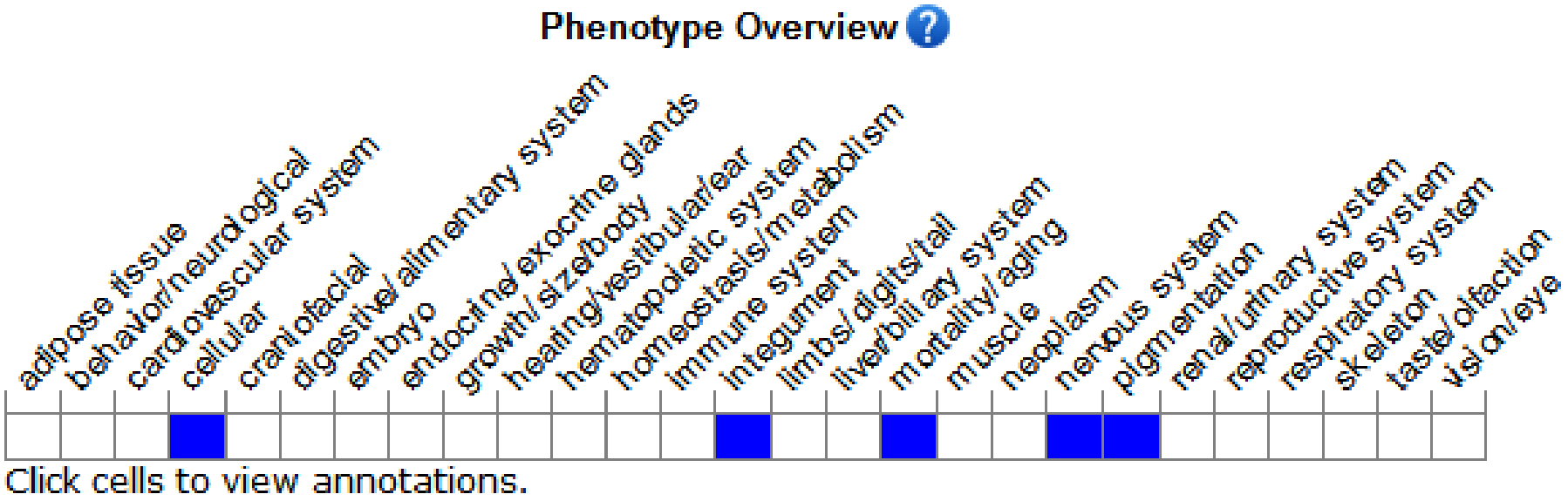


Genomic location distribution



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 targeted mutation exhibit exencephaly, neonatal lethality, and abnormal cerebellum morphology.

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

Tel: 025-5864 1534

集萃药康生物科技

GemPharmatech Co.,Ltd