Rgma Cas9-CKO Strategy

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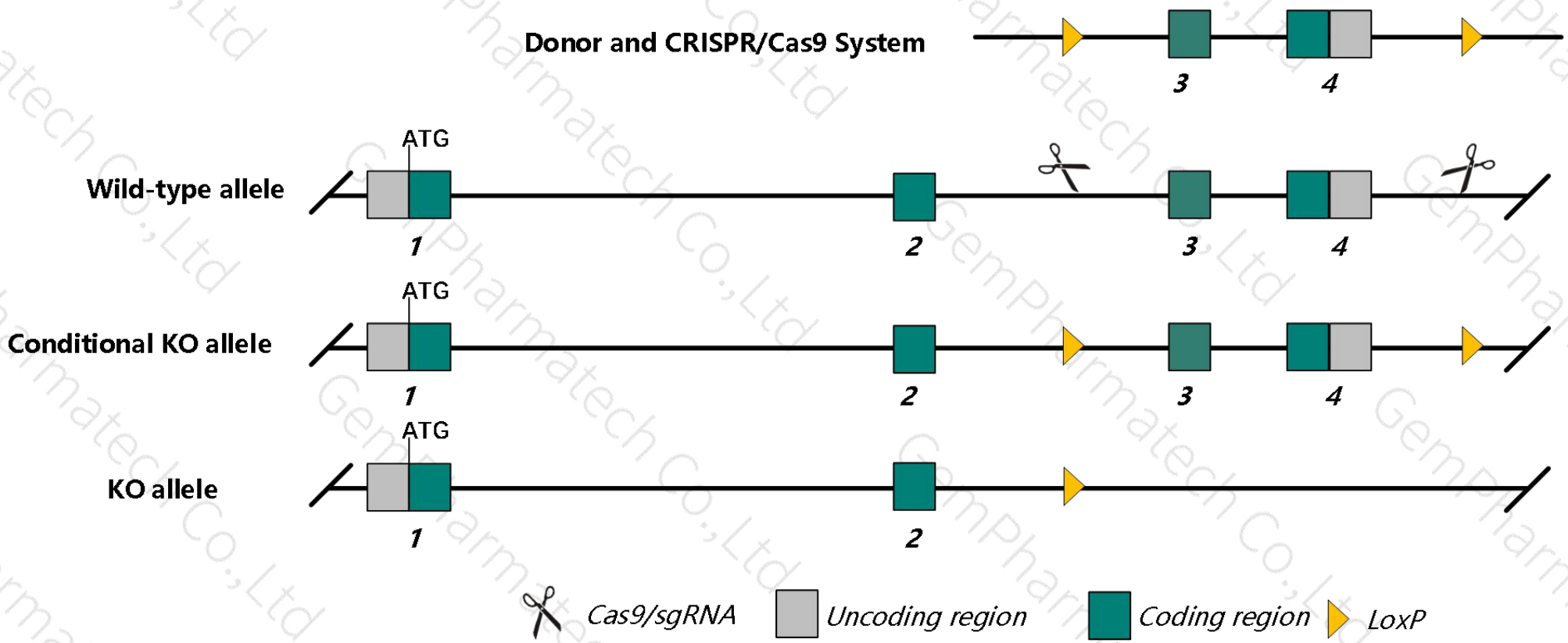
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Rgma* gene has 5 transcripts. According to the structure of *Rgma* gene, exon3-exon4 of *Rgma-201* (ENSMUST00000094312.11) transcript is recommended as the knockout region.The region contains most 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 *Rgma* 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,Inactivation of this locus results in impaired cephalic closure and subsequent exencephaly, both with incomplete penetrance. The retinal topography of the visual system is normal in homozygous mutant mice.

➢Transcript *Rgma*-205 may not be affected.

➢ The *Rgma* gene is located on the Chr7. 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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Rgma repulsive guidance molecule family member A [*Mus musculus* (house mouse)]

Gene ID: 244058, updated on 16-Dec-2019

Summary

Official Symbol	Rgma provided by MGI
Official Full Name	repulsive guidance molecule family member A provided by MGI
Primary source	MGI:MGI:2679262
See related	Ensembl:ENSMUSG00000070509
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	BC059072; C230063O06
Expression	Broad expression in whole brain E14.5 (RPKM 24.7), ovary adult (RPKM 23.2) and 23 other tissues See more
Orthologs	human all

Genomic context

Location: 7; 7 D1

See Rgma in Genome Data Viewer

Exon count: 5

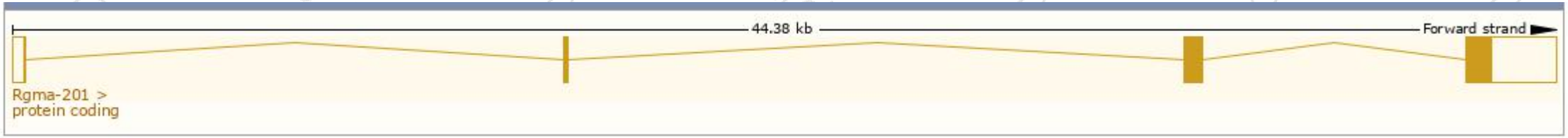
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	7	NC_000073.6 (73375520..73419899)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	7	NC_000073.5 (80520406..80564785)

Transcript information（Ensembl）

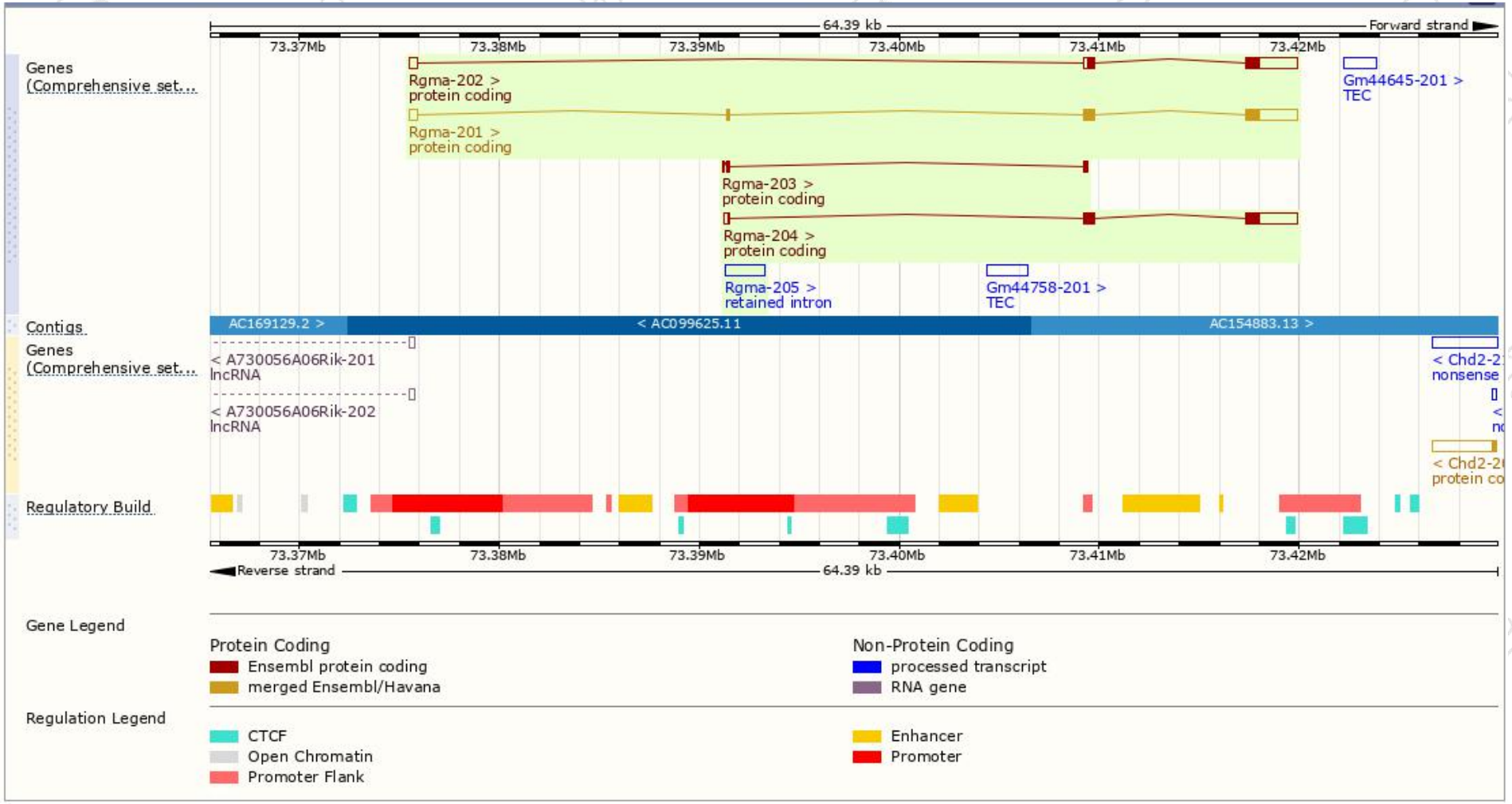
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID ▲	bp	Protein	Biotype	CCDS	UniProt	Flags
Rgma-201	ENSMUST00000094312.11	3586	454aa	Protein coding	CCDS21362	Q6PCX7	TSL:1 GENCODE basic APPRIS P2
Rgma-202	ENSMUST00000119206.2	3481	344aa	Protein coding	-	Q8C4C4	TSL:1 GENCODE basic
Rgma-203	ENSMUST00000128471.1	378	100aa	Protein coding	-	D3Z4W0	CDS 3' incomplete TSL:3
Rgma-204	ENSMUST00000139780.2	3371	438aa	Protein coding	-	Q6PCX7	TSL:1 GENCODE basic APPRIS ALT2
Rgma-205	ENSMUST00000206253.1	1972	No protein	Retained intron	-	-	TSL:NA

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

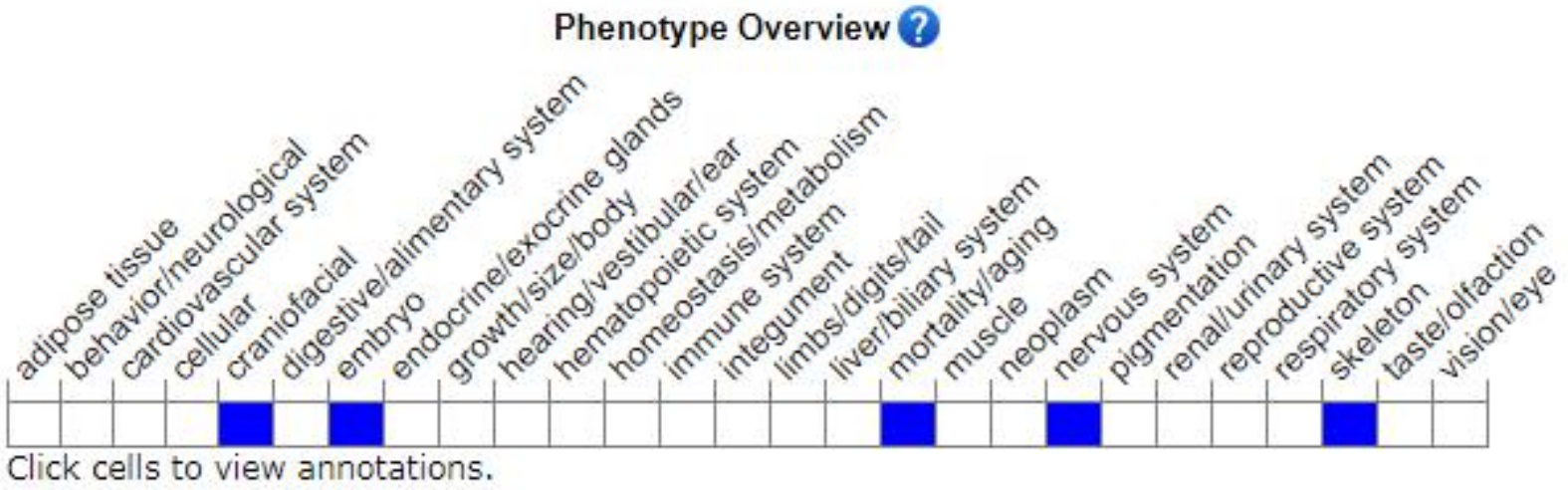


Genomic location distribution



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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