

# Exoc4 Cas9-CKO Strategy

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# **Project Overview**



Project Name Exoc4

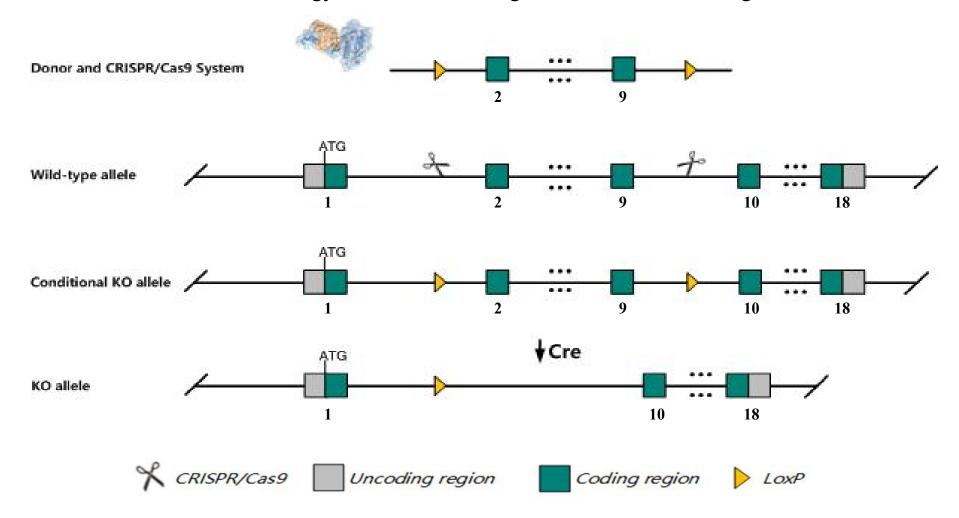
Project type Cas9-CKO

Strain background C57BL/6JGpt

## **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Exoc4* gene. The schematic diagram is as follows:



### **Technical routes**



The *Exoc4* gene has 10 transcripts. According to the structure of *Exoc4* gene, exon2-exon9 of *Exoc4-201* (ENSMUST00000052266.14) transcript is recommended as the knockout region. The region contains 1334bp coding sequence.

Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Exoc4* 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.





According to the existing MGI data, Mice homozygous for disruptions in this gene display embryonic abnormatlities. Gastrulation is not completed and mesoderm formation is abnormal. Death occurs before E10.5.

The *Exoc4* gene is located on the Chr6. 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



#### Exoc4 exocyst complex component 4 [Mus musculus (house mouse)]

Gene ID: 20336, updated on 19-Mar-2019

#### Summary

☆ ?

Official Symbol Exoc4 provided by MGI

Official Full Name exocyst complex component 4 provided by MGI

Primary source MGI:MGI:1096376

See related Ensembl:ENSMUSG00000029763

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 C78892, Sec8, Sec8l1

Expression Ubiquitous expression in CNS E14 (RPKM 5.5), CNS E11.5 (RPKM 5.2) and 28 other tissuesSee more

Orthologs <u>human</u> all

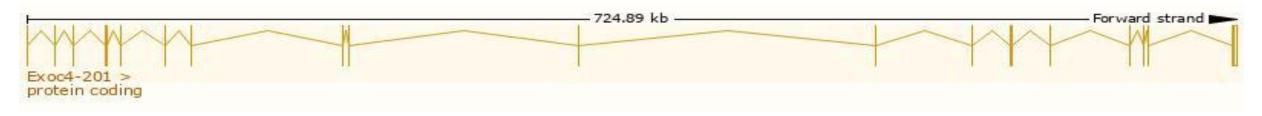
## Transcript information Ensembl



The gene has 10 transcripts, all transcripts are shown below:

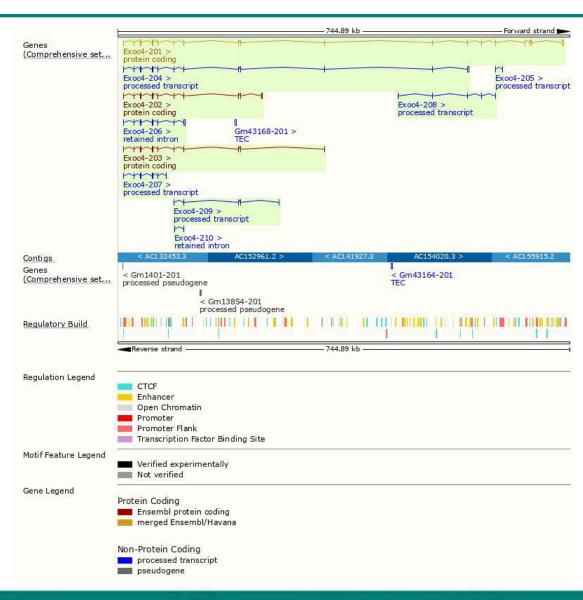
| Name      | Transcript ID         | bp   | Protein      | Biotype              | CCDS      | UniProt        | Flags                         |
|-----------|-----------------------|------|--------------|----------------------|-----------|----------------|-------------------------------|
| Exoc4-201 | ENSMUST00000052266.14 | 4824 | 975aa        | Protein coding       | CCDS19987 | <u>O35382</u>  | TSL:1 GENCODE basic APPRIS P1 |
| Exoc4-202 | ENSMUST00000090381.10 | 2598 | <u>522aa</u> | Protein coding       | CCDS85028 | Q9CXE1         | TSL:1 GENCODE basic           |
| Exoc4-203 | ENSMUST00000115080.1  | 2369 | 506aa        | Protein coding       | -         | Q8C391         | TSL:1 GENCODE basic           |
| Exoc4-204 | ENSMUST00000132842.7  | 3686 | No protein   | Processed transcript | 22        | 100            | TSL:1                         |
| Exoc4-207 | ENSMUST00000139132.1  | 1060 | No protein   | Processed transcript | -         | 1781           | TSL:1                         |
| Exoc4-209 | ENSMUST00000152835.1  | 932  | No protein   | Processed transcript | -         |                | TSL:3                         |
| Exoc4-208 | ENSMUST00000143190.1  | 641  | No protein   | Processed transcript | -         | 13 <b>2</b> -0 | TSL:5                         |
| Exoc4-205 | ENSMUST00000134653.1  | 384  | No protein   | Processed transcript | 82        | 828            | TSL:3                         |
| Exoc4-206 | ENSMUST00000137043.7  | 4225 | No protein   | Retained intron      | -         | 1723           | TSL:1                         |
| Exoc4-210 | ENSMUST00000156207.1  | 411  | No protein   | Retained intron      |           | 6.00           | TSL:3                         |

The strategy is based on the design of *Exoc4-201* transcript, The transcription is shown below



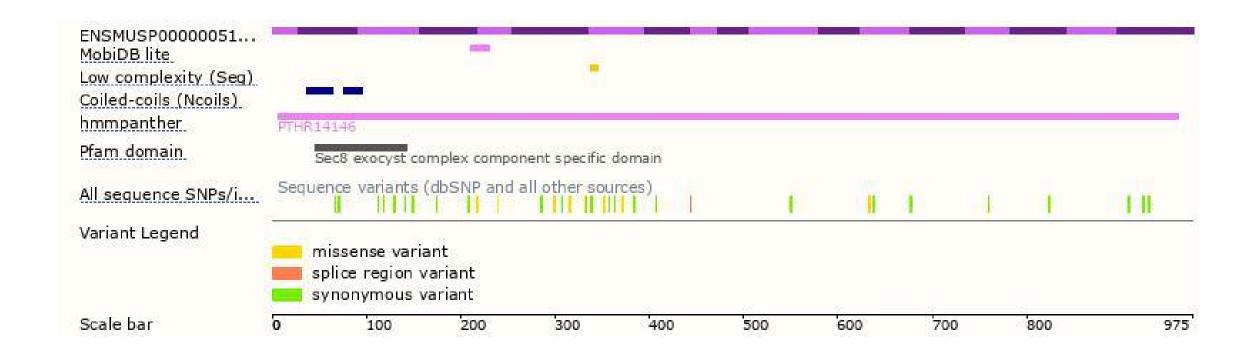
## Genomic location distribution





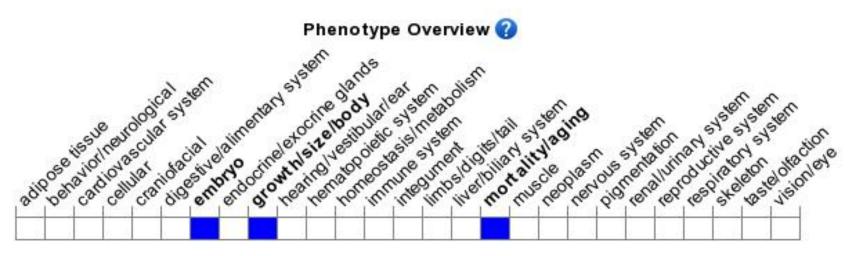
### Protein domain





# 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 disruptions in this gene display embryonic abnormatlities. Gastrulation is not completed and mesoderm formation is abnormal. Death occurs before E10.5.



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





