Orc6 Cas9-CKO Strategy

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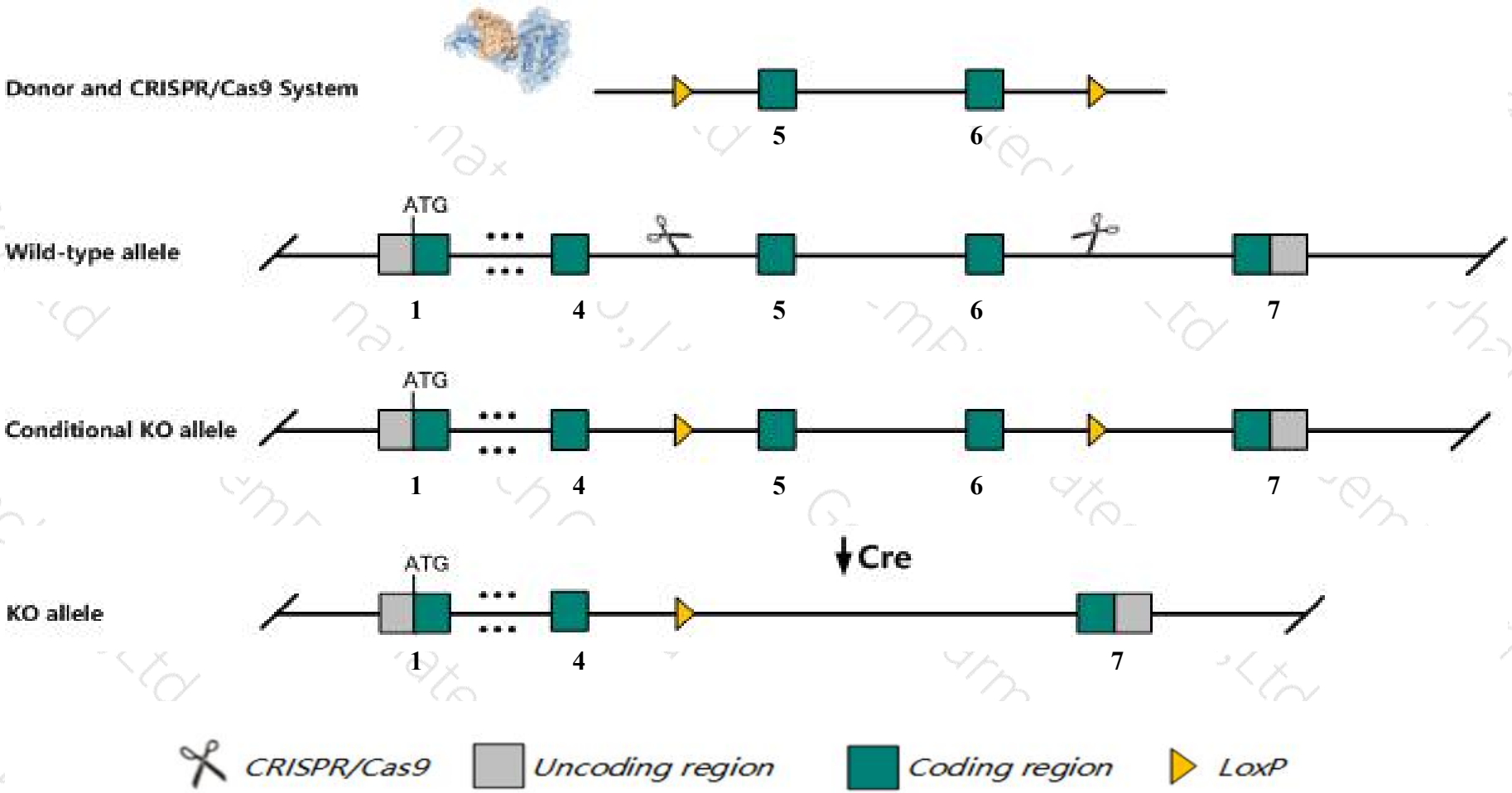
Project Overview

Project Name	***Orc6***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Orc6* gene. The schematic diagram is as follows:



Technical routes

- The *Orc6* gene has 7 transcripts. According to the structure of *Orc6* gene, exon5-exon6 of *Orc6-201* (ENSMUST00000034132.12) transcript is recommended as the knockout region.The region contains 215bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Orc6* 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 show complete embryonic lethality before implantation associated with abnormal morula morphology, increased cell death, and failure of blastocyst formation.
- The *Orc6* gene is located on the Chr8. 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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Orc6 origin recognition complex, subunit 6 [Mus musculus (house mouse)]

Gene ID: 56452, updated on 13-Mar-2020

Summary

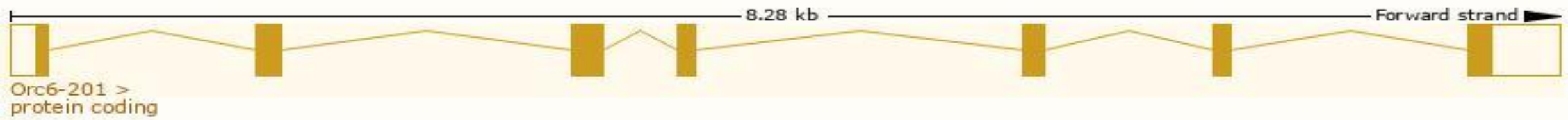
Official Symbol	Orc6 provided by MGI
Official Full Name	origin recognition complex, subunit 6 provided by MGI
Primary source	MGI:MGI:1929285
See related	Ensembl:ENSMUSG00000031697
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	6720420I10Rik, Orc6l
Expression	Broad expression in liver E14 (RPKM 33.8), CNS E11.5 (RPKM 28.8) and 21 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

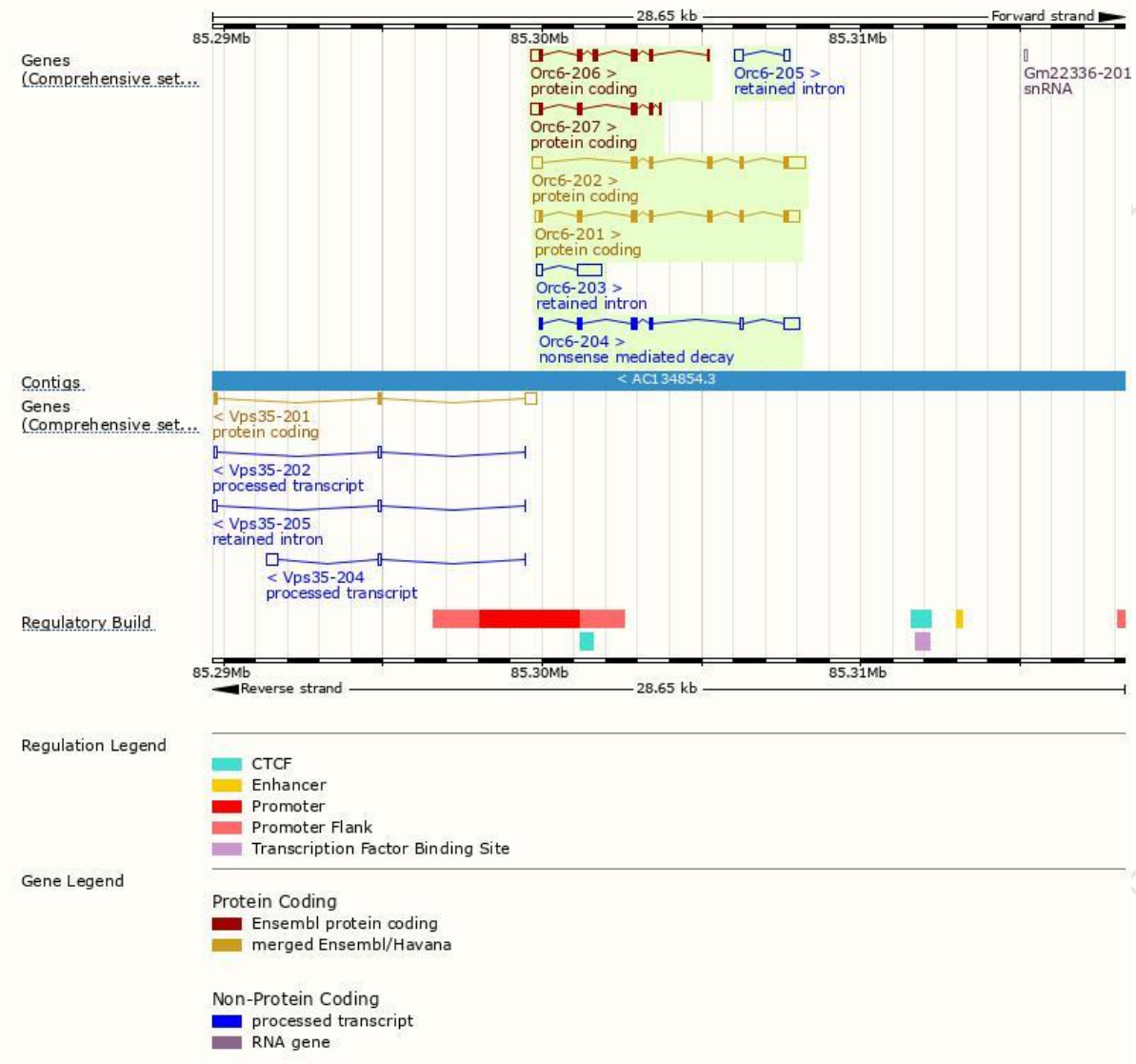
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Orc6-202	ENSMUST00000170141.2	1466	186aa	Protein coding	CCDS52623	Q3TQX1	TSL:1 GENCODE basic
Orc6-201	ENSMUST00000034132.12	1287	262aa	Protein coding	CCDS22497	Q66JV6	TSL:1 GENCODE basic APPRIS P1
Orc6-206	ENSMUST00000211396.1	887	197aa	Protein coding	-	A0A1B0GRE5	CDS 3' incomplete TSL:3
Orc6-207	ENSMUST00000211597.1	777	160aa	Protein coding	-	A0A1B0GSE2	CDS 3' incomplete TSL:3
Orc6-204	ENSMUST00000210146.1	1041	156aa	Nonsense mediated decay	-	A0A1B0GR56	TSL:1
Orc6-203	ENSMUST00000209733.1	921	No protein	Retained intron	-	-	TSL:2
Orc6-205	ENSMUST00000210458.1	461	No protein	Retained intron	-	-	TSL:2

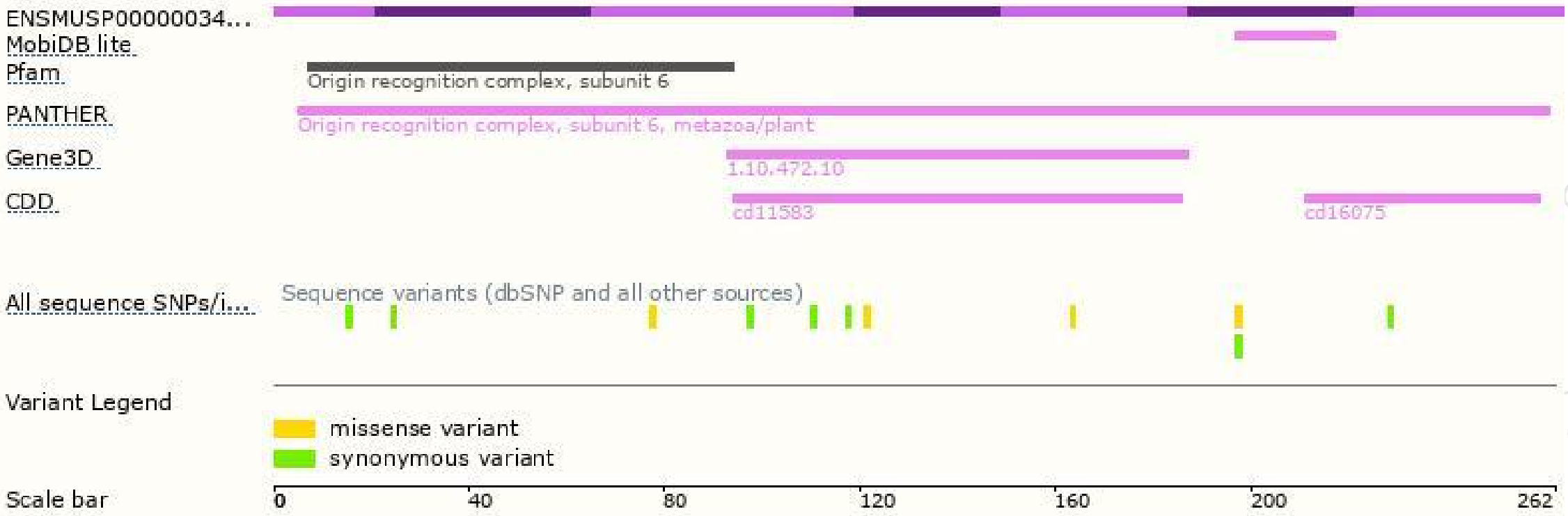
The strategy is based on the design of *Orc6-201* transcript,the transcription is shown below:



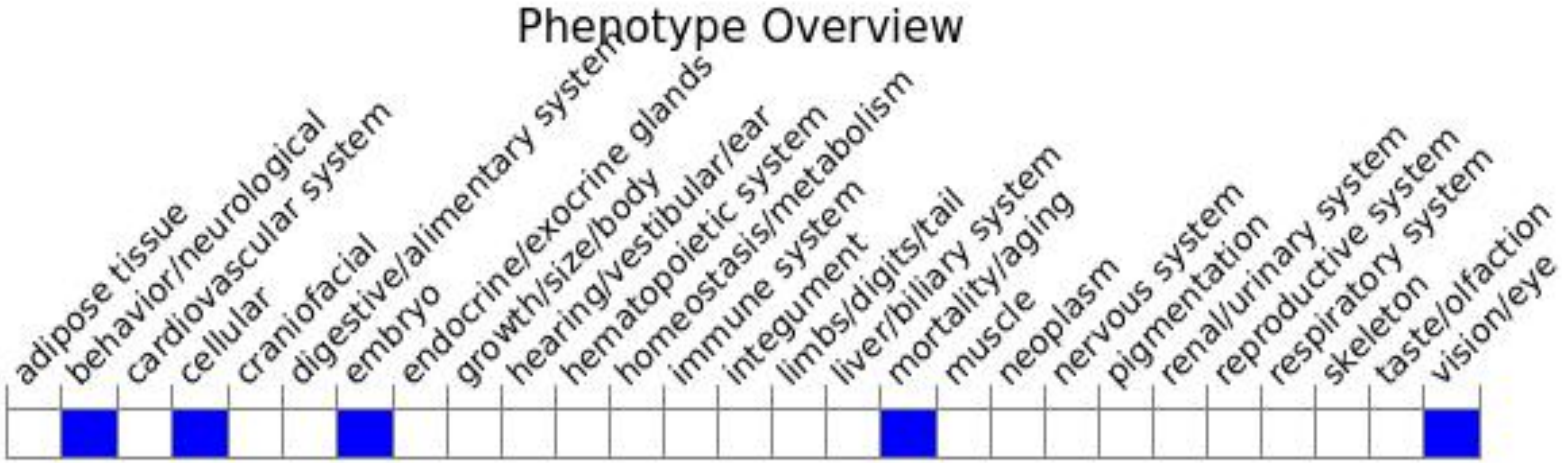
Genomic location distribution



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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