

C2cd4d Cas9-CKO Strategy

Designer: Daohua Xu

Reviewer: Xueting Zhang

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Project Overview

Project Name

C2cd4d

Project type

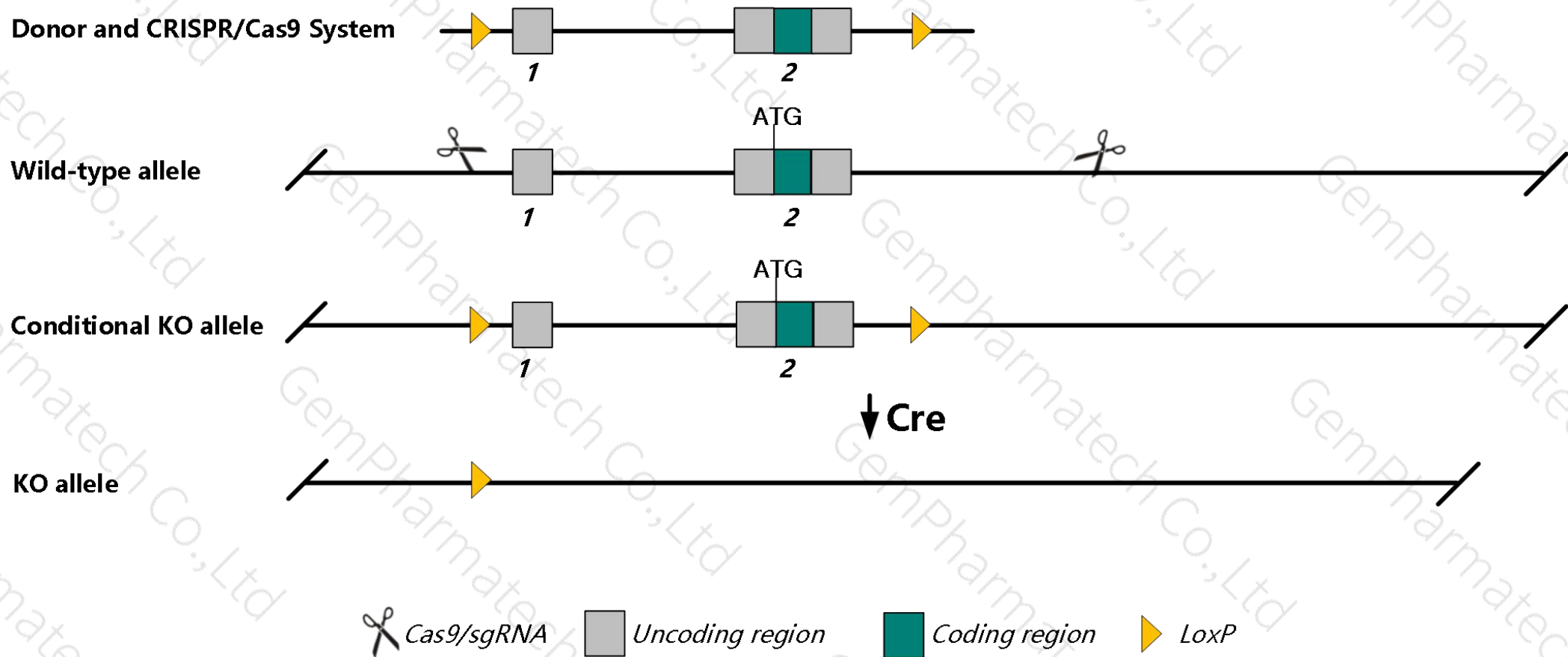
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *C2cd4d* gene. The schematic diagram is as follows:



- The *C2cd4d* gene has 2 transcripts. According to the structure of *C2cd4d* gene, exon1-exon2 of *C2cd4d-201*(ENSMUST00000169433.2) transcript is recommended as the knockout region. The region contains all 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 *C2cd4d* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 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.

- The KO region contains functional region of the *Gm36070* gene. Knockout the region may affect the function of *Gm36070* gene.
- The *C2cd4d* gene is located on the Chr3. 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)

C2cd4d C2 calcium-dependent domain containing 4D [Mus musculus (house mouse)]

Gene ID: 271944, updated on 13-Mar-2020

Summary



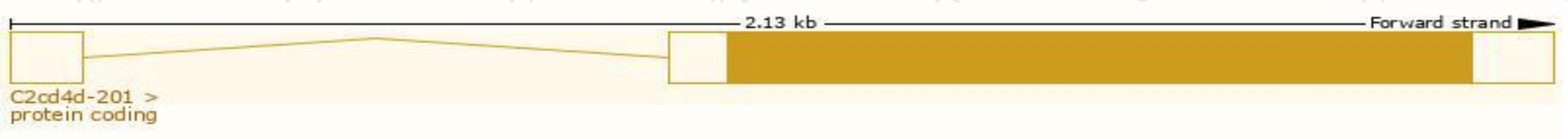
Official Symbol	C2cd4d provided by MGI
Official Full Name	C2 calcium-dependent domain containing 4D provided by MGI
Primary source	MGI:MGI:2685505
See related	Ensembl:ENSMUSG00000091648
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gm659
Expression	Biased expression in thymus adult (RPKM 10.2), ovary adult (RPKM 3.3) and 13 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

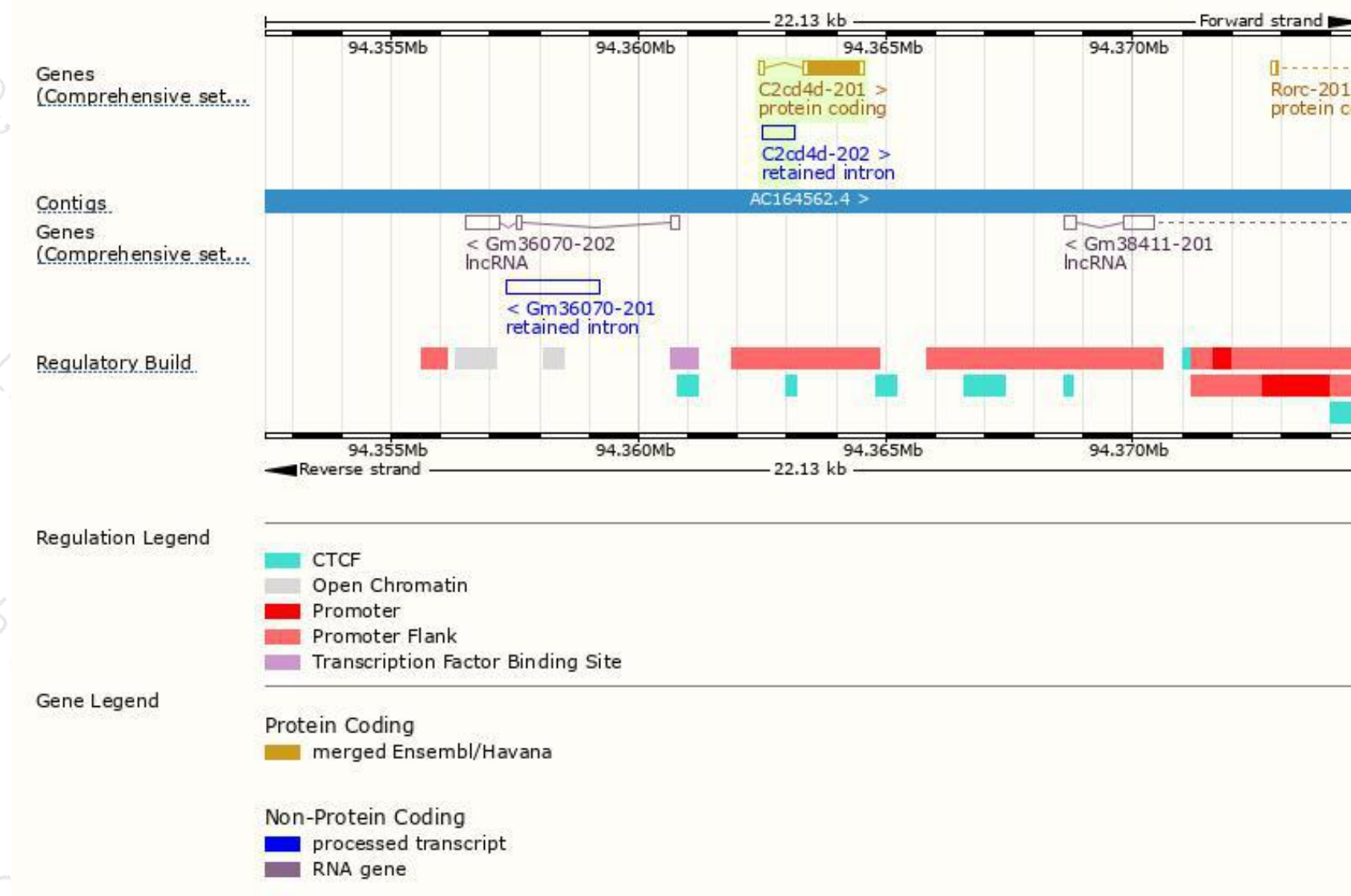
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
C2cd4d-201	ENSMUST00000169433.2	1321	341aa	Protein coding	CCDS50983	P0CG09	TSL:3 GENCODE basic APPRIS P1
C2cd4d-202	ENSMUST00000197526.1	662	No protein	Retained intron	-	-	TSL:NA

The strategy is based on the design of *C2cd4d-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

Tel: 025-5864 1534

