

C2cd5 Cas9-CKO Strategy

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Project Overview



Project Name C2cd5

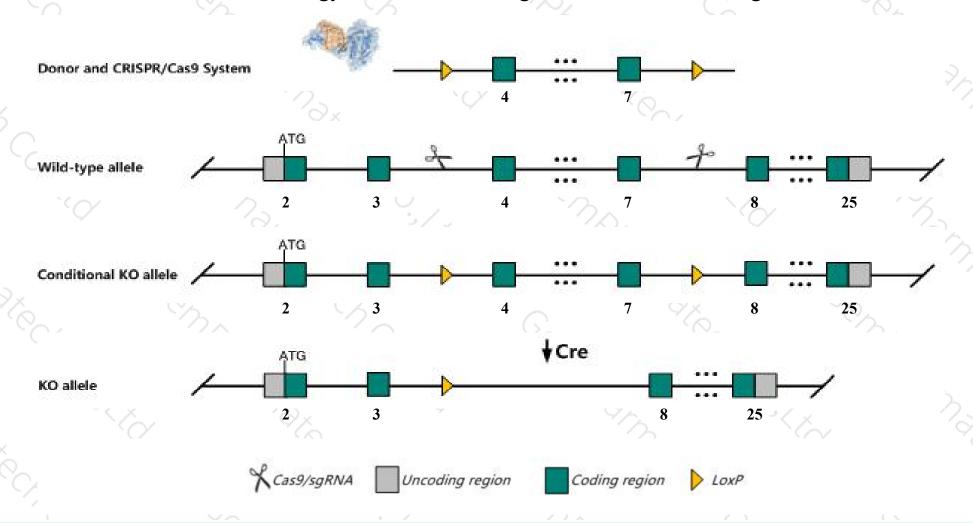
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the C2cd5 gene. The schematic diagram is as follows:



Technical routes



- ➤ The C2cd5 gene has 17 transcripts. According to the structure of C2cd5 gene, exon4-exon7 of C2cd5-203(ENSMUST00000171349.7) transcript is recommended as the knockout region. The region contains 623bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *C2cd5* 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.

Notice



- > The C2cd5 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)



C2cd5 C2 calcium-dependent domain containing 5 [Mus musculus (house mouse)]

Gene ID: 74741, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol C2cd5 provided by MGI

Official Full Name C2 calcium-dependent domain containing 5 provided by MGI

Primary source MGI:MGI:1921991

See related Ensembl: ENSMUSG00000030279

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 5730419109Rik, C030008B15Rik, CDP138

Expression Broad expression in CNS E18 (RPKM 8.8), CNS E14 (RPKM 8.4) and 25 other tissuesSee more

Orthologs <u>human all</u>

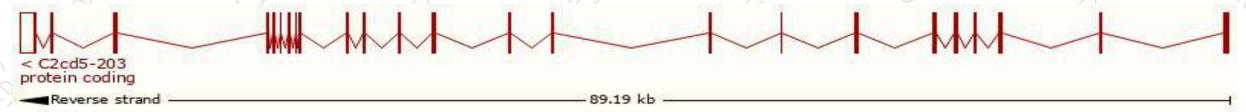
Transcript information (Ensembl)



The gene has 17 transcripts, all transcripts are shown below:

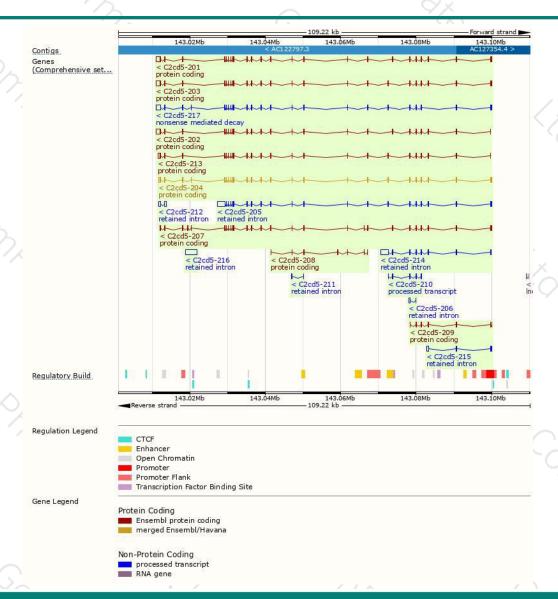
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
C2cd5-203	ENSMUST00000171349.7	4292	999aa	Protein coding	CCDS51951	E9Q026	TSL:5 GENCODE basic APPRIS ALT1
C2cd5-201	ENSMUST00000087485.6	4265	990aa	Protein coding	CCDS51953	Q7TPS5	TSL:5 GENCODE basic
C2cd5-202	ENSMUST00000111758.8	4062	990aa	Protein coding	CCDS51953	Q7TPS5	TSL:1 GENCODE basic
C2cd5-213	ENSMUST00000204655.2	3526	999aa	Protein coding	CCDS51951	E9Q026	TSL:5 GENCODE basic APPRIS ALT1
C2cd5-204	ENSMUST00000203187.2	3494	1016aa	Protein coding	CCDS51952	Q7TPS5	TSL:1 GENCODE basic APPRIS P4
C2cd5-207	ENSMUST00000203673.2	3332	1052aa	Protein coding	-	A0A0N4SW93	TSL:5 GENCODE basic APPRIS ALT1
C2cd5-209	ENSMUST00000204140.1	771	232aa	Protein coding	-	A0A0N4SVN0	CDS 3' incomplete TSL:2
C2cd5-208	ENSMUST00000204043.1	543	<u>181aa</u>	Protein coding	-	A0A0N4SW58	CDS 5' and 3' incomplete TSL:5
C2cd5-217	ENSMUST00000205119.2	4149	<u>70aa</u>	Nonsense mediated decay	-	A0A0N4SW69	TSL:1
C2cd5-210	ENSMUST00000204160.1	685	No protein	Processed transcript	-	10-	TSL:3
C2cd5-205	ENSMUST00000203349.2	4584	No protein	Retained intron	2	828	TSL:1
C2cd5-214	ENSMUST00000204967.2	3122	No protein	Retained intron	8	-	TSL:1
C2cd5-216	ENSMUST00000205117.1	2993	No protein	Retained intron	- 1	-	TSL:NA
C2cd5-212	ENSMUST00000204635.1	909	No protein	Retained intron	-	878	TSL:1
C2cd5-215	ENSMUST00000205079.1	836	No protein	Retained intron	-		TSL:1
C2cd5-206	ENSMUST00000203537.1	573	No protein	Retained intron	2		TSL:3
C2cd5-211	ENSMUST00000204233.1	359	No protein	Retained intron	-	8578	TSL:3

The strategy is based on the design of *C2cd5-203* transcript, the transcription is shown below:



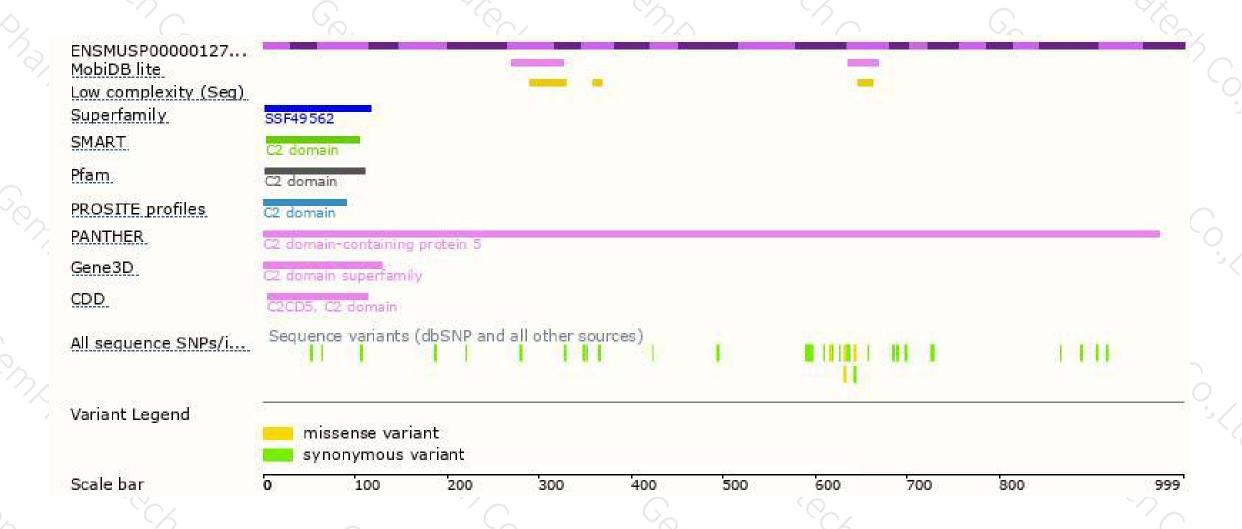
Genomic location distribution





Protein domain







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

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