

Dnajc6 Cas9-CKO Strategy

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Reviewer: JiaYu

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Project Overview



Project Name Dnajc6

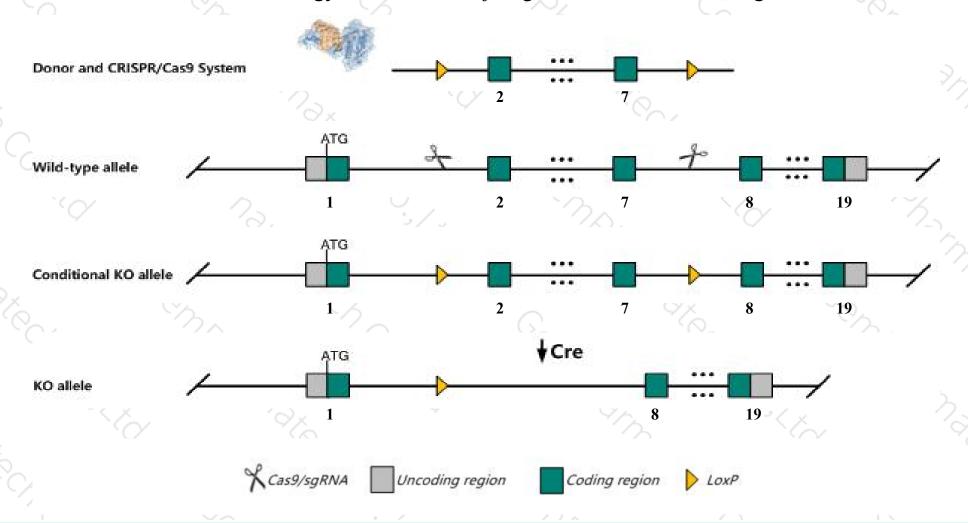
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Dnajc6* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Dnajc6* gene has 8 transcripts. According to the structure of *Dnajc6* gene, exon2-exon7 of *Dnajc6*205(ENSMUST00000106933.1) transcript is recommended as the knockout region. The region contains 802bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Dnajc6* 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



- > According to the existing MGI data,mice homozygous and heterozygous for a knock-out allele exhibit postnatal lethality and decreased body weight with homozygotes exhibiting decreased synpatic vesicle recycling.
- > The *Dnajc6* gene is located on the Chr4. 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)



Dnajc6 DnaJ heat shock protein family (Hsp40) member C6 [Mus musculus (house mouse)]

Gene ID: 72685, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Dnajc6 provided by MGI

Official Full Name DnaJ heat shock protein family (Hsp40) member C6 provided by MGI

Primary source MGI:MGI:1919935

See related Ensembl: ENSMUSG00000028528

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 2810027M23Rik, mKIAA0473

Expression Biased expression in frontal lobe adult (RPKM 27.2), cortex adult (RPKM 25.9) and 7 other tissuesSee more

Orthologs <u>human all</u>

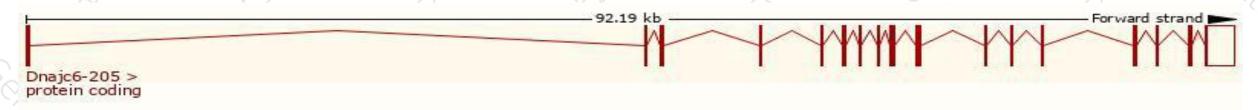
Transcript information (Ensembl)



The gene has 8 transcripts, all transcripts are shown below:

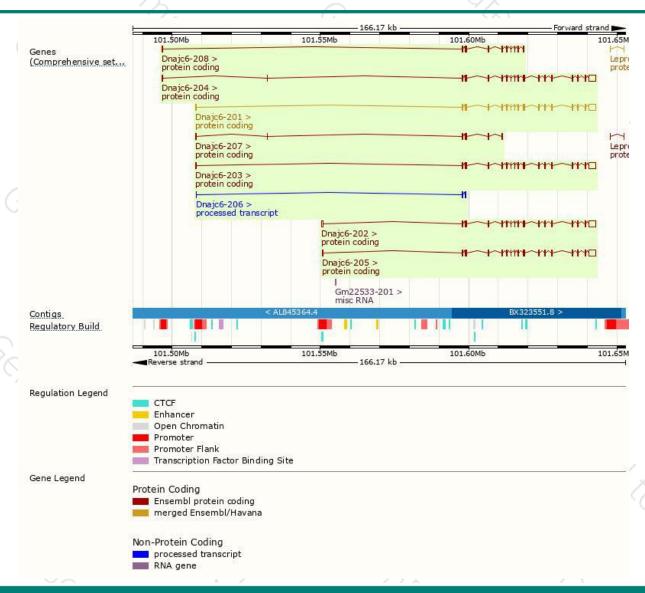
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST00000094953.10	5322	900aa	Protein coding	CCDS18395	Q80TZ3	TSL:1 GENCODE basic APPRIS P3
ENSMUST00000038207.11	5258	<u>938aa</u>	Protein coding	CCDS51237	Q80TZ3	TSL:1 GENCODE basic APPRIS ALT2
ENSMUST00000106933.1	5191	968aa	Protein coding	CCDS51238	Q80TZ3	TSL:1 GENCODE basic APPRIS ALT2
ENSMUST00000106929.9	5128	900aa	Protein coding	CCDS18395	Q80TZ3	TSL:1 GENCODE basic APPRIS P3
ENSMUST00000106930.7	5127	900aa	Protein coding	CCDS18395	Q80TZ3	TSL:1 GENCODE basic APPRIS P3
ENSMUST00000154120.8	1569	<u>514aa</u>	Protein coding	170	B1B0B7	CDS 3' incomplete TSL:5
ENSMUST00000149047.7	872	<u>194aa</u>	Protein coding	:	B1B0B8	CDS 3' incomplete TSL:3
ENSMUST00000146489.1	458	No protein	Processed transcript	122	(4)	TSL:3
	ENSMUST00000094953.10 ENSMUST00000038207.11 ENSMUST00000106933.1 ENSMUST00000106929.9 ENSMUST00000106930.7 ENSMUST00000154120.8 ENSMUST00000149047.7	ENSMUST000000106933.1 5191 ENSMUST00000106933.1 5191 ENSMUST00000106929.9 5128 ENSMUST00000106930.7 5127 ENSMUST00000154120.8 1569 ENSMUST00000149047.7 872	ENSMUST00000094953.10 5322 900aa ENSMUST00000038207.11 5258 938aa ENSMUST00000106933.1 5191 968aa ENSMUST00000106929.9 5128 900aa ENSMUST00000106930.7 5127 900aa ENSMUST00000154120.8 1569 514aa ENSMUST00000149047.7 872 194aa	ENSMUST00000094953.10 5322 900aa Protein coding ENSMUST00000038207.11 5258 938aa Protein coding ENSMUST00000106933.1 5191 968aa Protein coding ENSMUST00000106929.9 5128 900aa Protein coding ENSMUST00000106930.7 5127 900aa Protein coding ENSMUST00000154120.8 1569 514aa Protein coding ENSMUST00000149047.7 872 194aa Protein coding	ENSMUST00000094953.10 5322 900aa Protein coding CCDS18395 ENSMUST00000038207.11 5258 938aa Protein coding CCDS51237 ENSMUST00000106933.1 5191 968aa Protein coding CCDS51238 ENSMUST00000106929.9 5128 900aa Protein coding CCDS18395 ENSMUST00000106930.7 5127 900aa Protein coding CCDS18395 ENSMUST00000154120.8 1569 514aa Protein coding - ENSMUST00000149047.7 872 194aa Protein coding -	ENSMUST00000094953.10 5322 900aa Protein coding CCDS18395 Q80TZ3 ENSMUST00000038207.11 5258 938aa Protein coding CCDS51237 Q80TZ3 ENSMUST00000106933.1 5191 968aa Protein coding CCDS51238 Q80TZ3 ENSMUST00000106929.9 5128 900aa Protein coding CCDS18395 Q80TZ3 ENSMUST00000106930.7 5127 900aa Protein coding CCDS18395 Q80TZ3 ENSMUST000000154120.8 1569 514aa Protein coding - B1B0B7 ENSMUST000000149047.7 872 194aa Protein coding - B1B0B8

The strategy is based on the design of *Dnajc6-205* transcript, the transcription is shown below:



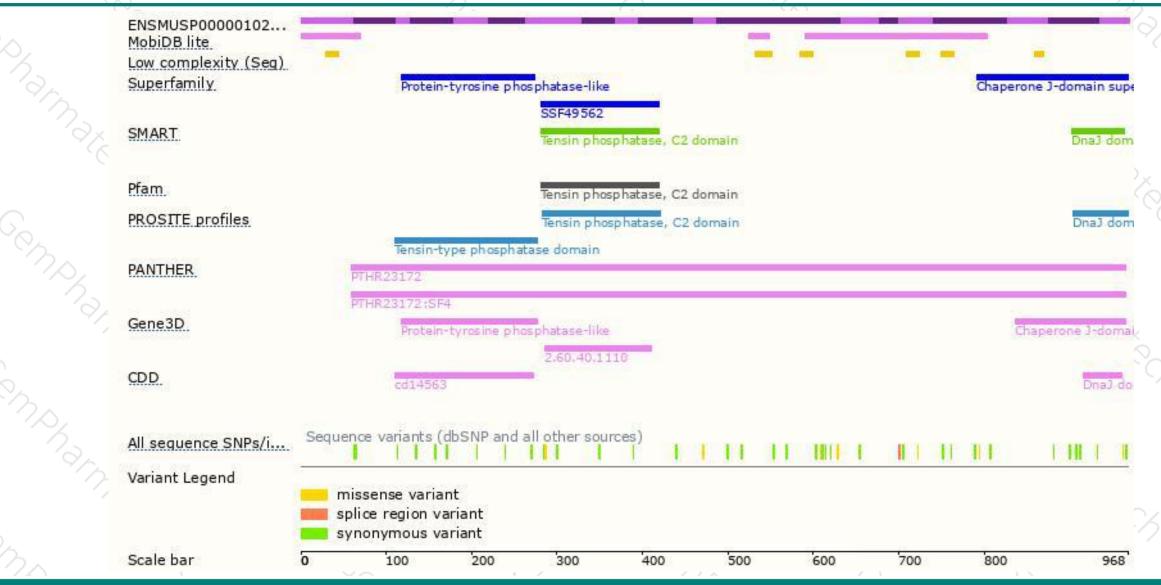
Genomic location distribution





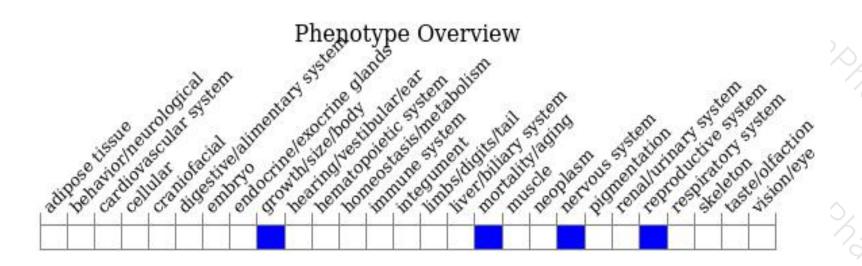
Protein domain





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 and heterozygous for a knock-out allele exhibit postnatal lethality and decreased body weight with homozygotes exhibiting decreased synpatic vesicle recycling.



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

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