

Drp2 Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Daohua Xu

Design Date: 2020-7-27

Project Overview



Project Name

Drp2

Project type

Cas9-CKO

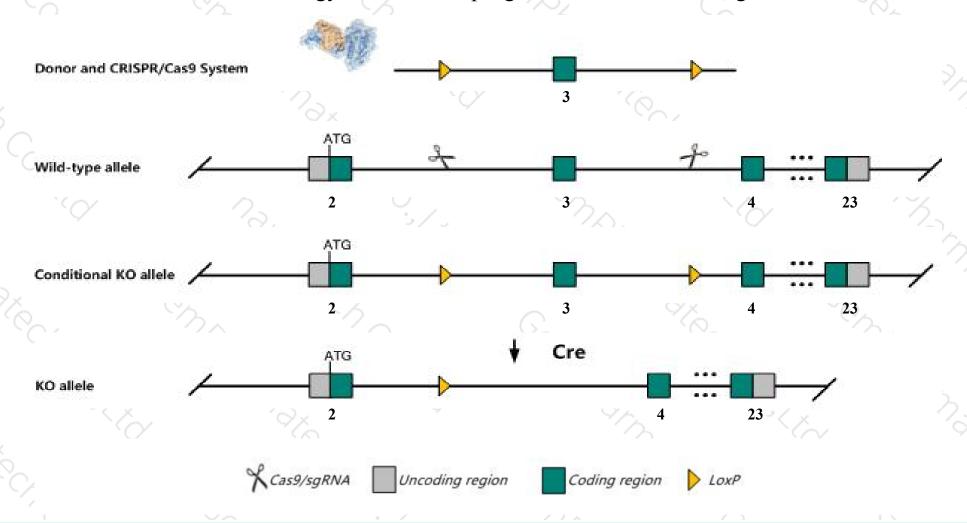
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Drp2* gene. The schematic diagram is as follows:



Technical routes



- > The *Drp2* gene has 6 transcripts. According to the structure of *Drp2* gene, exon3 of *Drp2-206*(ENSMUST00000153424.7) transcript is recommended as the knockout region. The region contains 164bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Drp2* 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 for a conditional allele activated in peripheral nerves exhibit abnormal Scwann cell morphology, supernumerary Schwann cell, abnormal myelin sheath morphology and hypermyelination.
- > The *Drp2* gene is located on the ChrX. 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)



Drp2 dystrophin related protein 2 [Mus musculus (house mouse)]

Gene ID: 13497, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Drp2 provided by MGI

Official Full Name dystrophin related protein 2 provided by MGI

Primary source MGI:MGI:107432

See related Ensembl:ENSMUSG00000000223

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 AW495265, DRP-2

Expression Biased expression in frontal lobe adult (RPKM 10.1), cortex adult (RPKM 10.1) and 10 other tissuesSee more

Orthologs <u>human all</u>

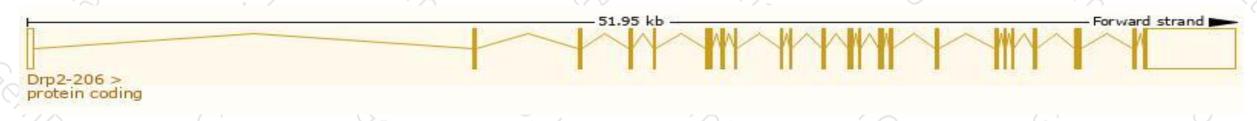
Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

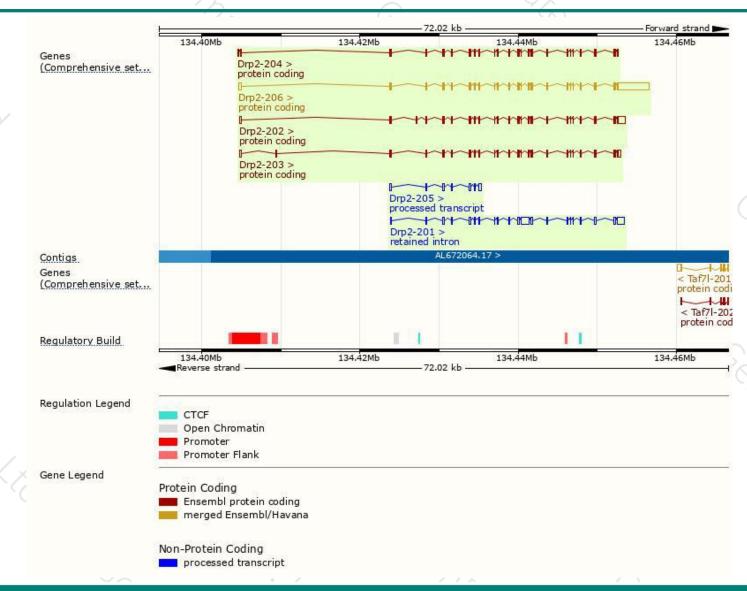
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Drp2-206	ENSMUST00000153424.7	7135	957aa	Protein coding	CCDS30394	Q05AA6	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Drp2-203	ENSMUST00000113226.1	3499	<u>957aa</u>	Protein coding	CCDS30394	Q05AA6	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Drp2-204	ENSMUST00000113228.7	3141	957aa	Protein coding	CCDS30394	Q05AA6	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Drp2-202	ENSMUST00000113224.8	4147	<u>981aa</u>	Protein coding		B1AV35	TSL:5 GENCODE basic
Drp2-205	ENSMUST00000142146.7	1423	No protein	Processed transcript	-	-	TSL:5
Drp2-201	ENSMUST00000000228.4	4389	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Drp2-206* transcript, the transcription is shown below:



Genomic location distribution





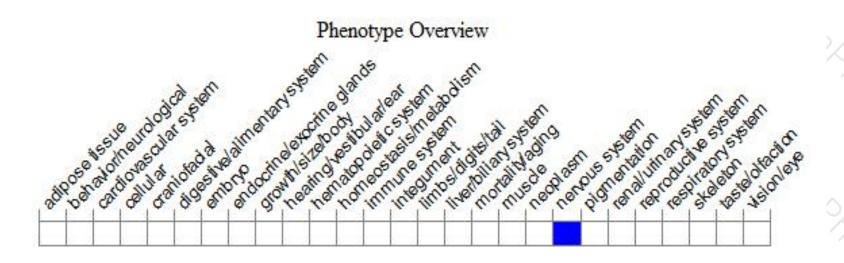
Protein domain





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 a conditional allele activated in peripheral nerves exhibit abnormal Scwann cell morphology, supernumerary Schwann cell, abnormal myelin sheath morphology and hypermyelination.



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

Tel: 025-5864 1534





