Drp2 Cas9-KO Strategy

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Reviewer: Daohua Xu

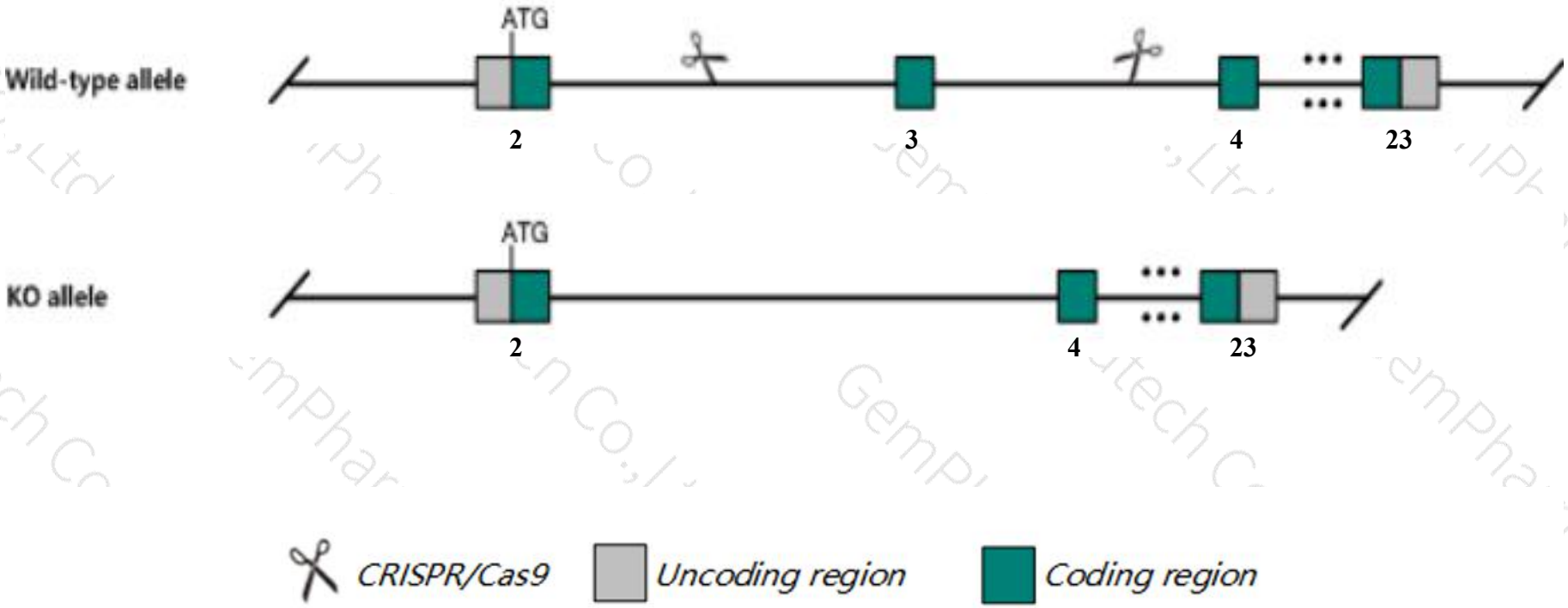
Design Date: 2020-7-27

Project Overview

Project Name	***Drp2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

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: CRISPR/Cas9 system 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.

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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

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

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

Summary

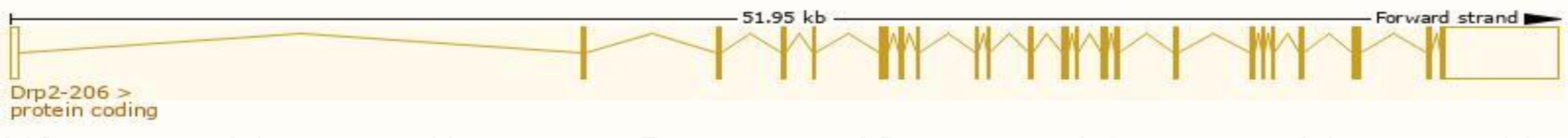
Official Symbol	Drp2 provided by MGI
Official Full Name	dystrophin related protein 2 provided byMGI
Primary source	MGI:MGI:107432
See related	Ensembl:ENSMUSG00000000223
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; 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	human all

Transcript information（Ensembl）

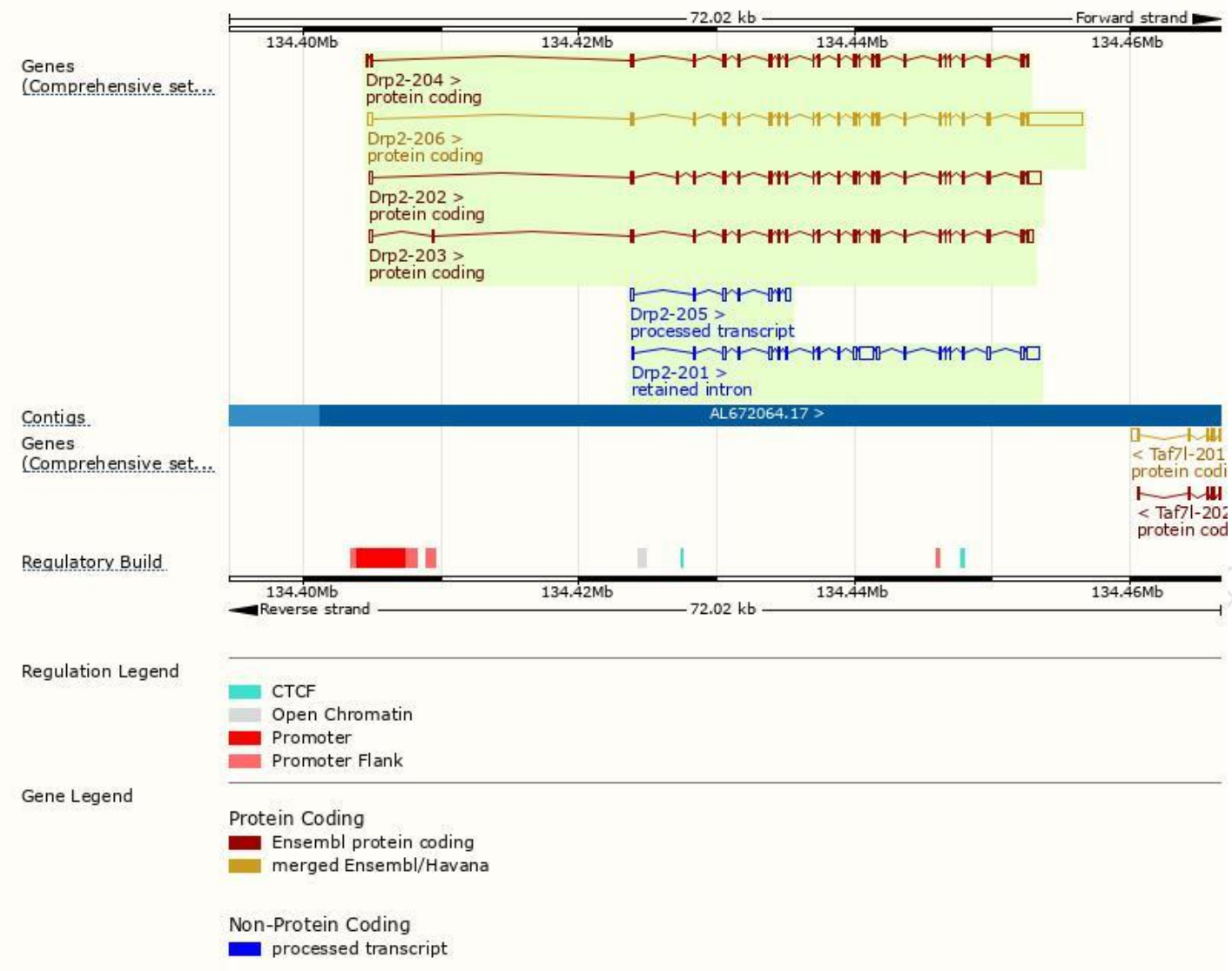
The gene has 6 transcripts,all transcripts are shown below:

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	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
Drp2-204	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	981aa	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

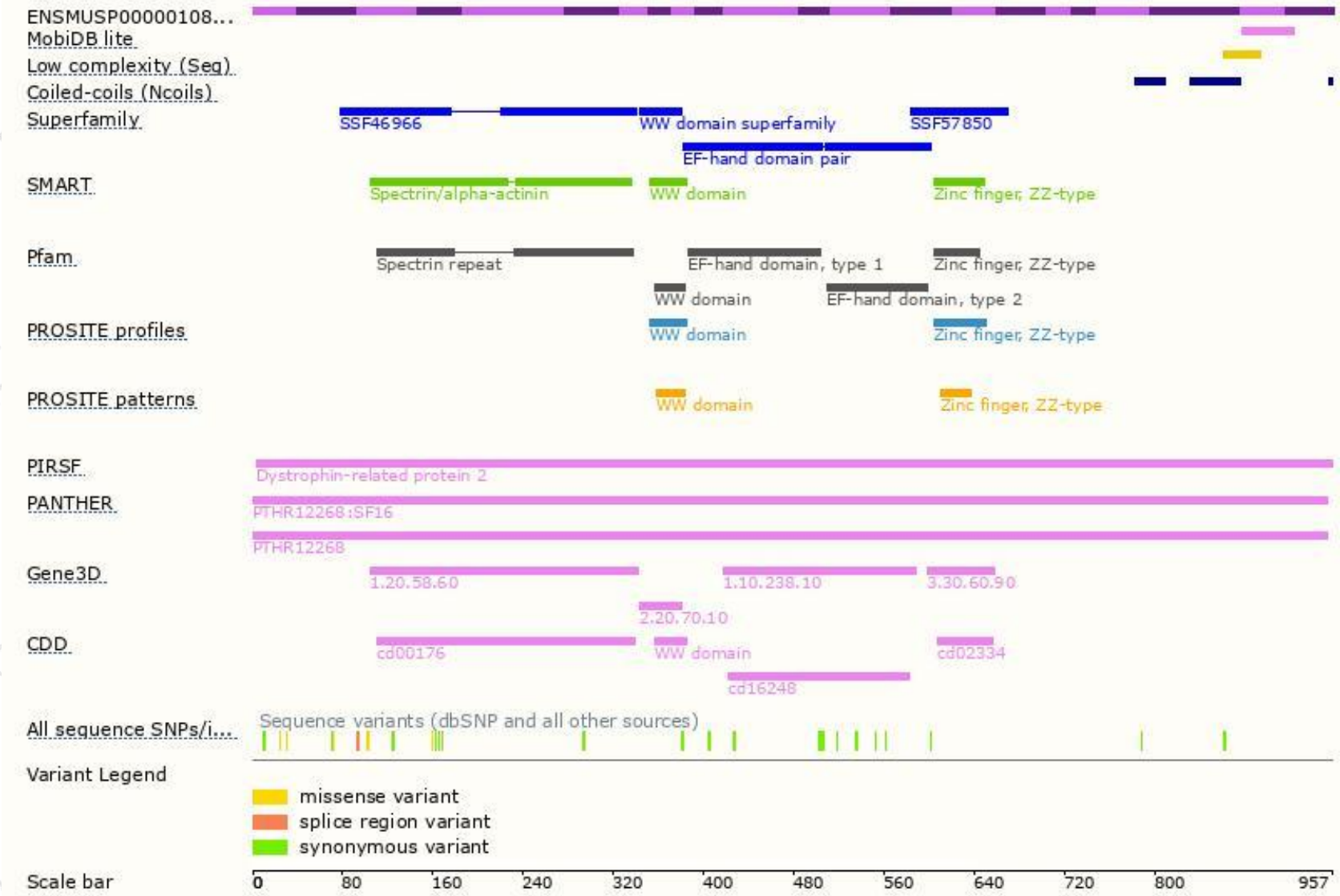
The strategy is based on the design of *Drp2-206* transcript,the transcription is shown below:



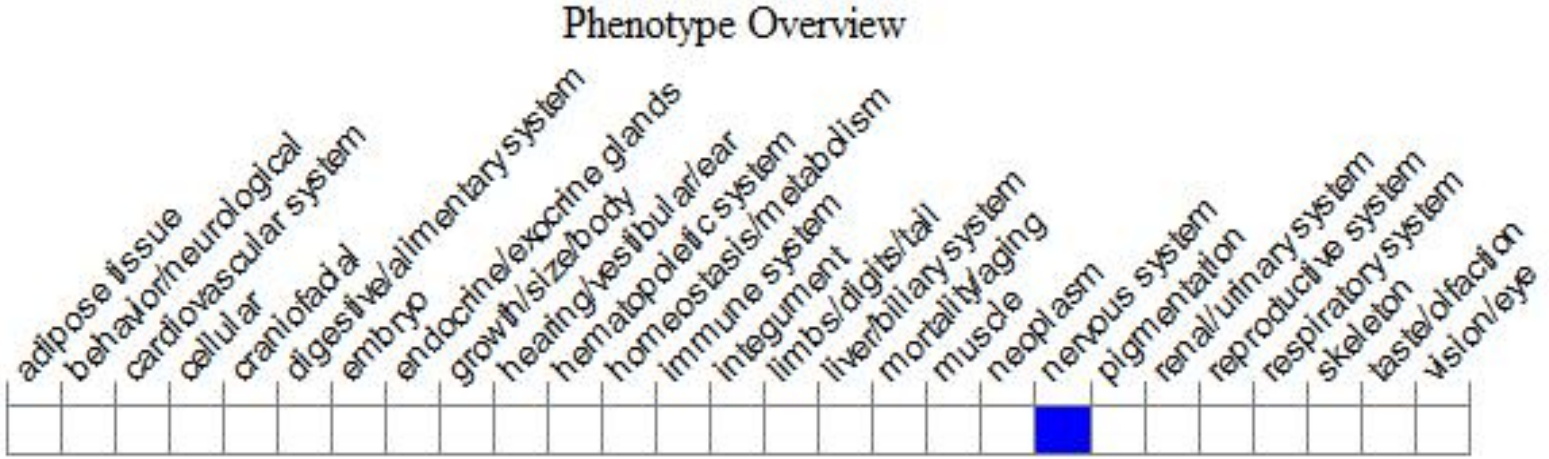
Genomic location distribution



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.

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

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