Rufy3 Cas9-KO Strategy

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Design Date: 2020-4-23

Project Overview

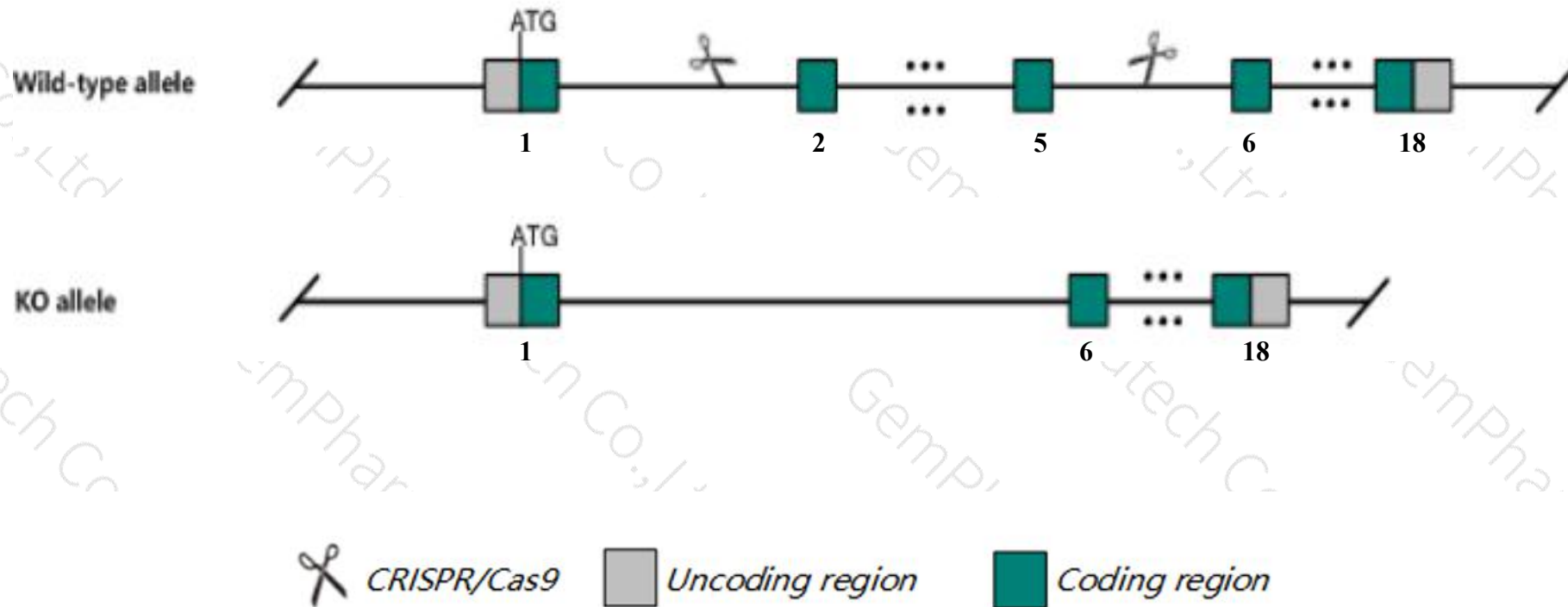|	
Project Name	*Rufy3*
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rufy3* gene. The schematic diagram is as follows:

Technical routes

➢ The *Rufy3* gene has 8 transcripts. According to the structure of *Rufy3* gene, exon2-exon5 of *Rufy3-205* (ENSMUST00000198965.4) transcript is recommended as the knockout region.The region contains 518bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Rufy3* gene. The brief process is as follows: CRISPR/Cas9 system

Notice

➢ According to the existing MGI data,the gene product is involved in regulating neuronal polarity and axon

  growth. homozygous ko leads to loss of neuronal polarity and causes neonatal lethality.

➢ The *Rufy3* gene is located on the Chr5. 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）

Rufy3 RUN and FYVE domain containing 3 [Mus musculus (house mouse)]

Gene ID: 52822, updated on 13-Mar-2020

Summary

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Official Symbol	Rufy3 provided by MGI
Official Full Name	RUN and FYVE domain containing 3 provided by MGI
Primary source	MGI:MGI:106484
See related	Ensembl:ENSMUSG00000029291
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	2810428M05Rik, 6330416M07Rik, AW455998, AW538594, D5Bwg0860e, Ripx, Rpipx
Expression	Broad expression in CNS E18 (RPKM 20.6), whole brain E14.5 (RPKM 18.1) and 18 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rufy3-201	ENSMUST00000031229.10	4049	469aa	Protein coding	CCDS19403	Q9D394	TSL:1 GENCODE basic
Rufy3-203	ENSMUST00000196894.4	3703	519aa	Protein coding	CCDS80323	Q9D394	TSL:1 GENCODE basic
Rufy3-205	ENSMUST00000198965.4	3536	669aa	Protein coding	CCDS80324	A0A0G2JFT8	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
Rufy3-207	ENSMUST00000199312.4	2182	487aa	Protein coding	CCDS80325	Q9D394	TSL:1 GENCODE basic
Rufy3-202	ENSMUST00000196686.1	2077	487aa	Protein coding	CCDS80325	Q9D394	TSL:1 GENCODE basic
Rufy3-206	ENSMUST00000199207.1	2722	No protein	Retained intron	-	-	TSL:2
Rufy3-204	ENSMUST00000197664.1	2336	No protein	Retained intron	-	-	TSL:NA
Rufy3-208	ENSMUST00000199956.1	619	No protein	Retained intron	-	-	TSL:3

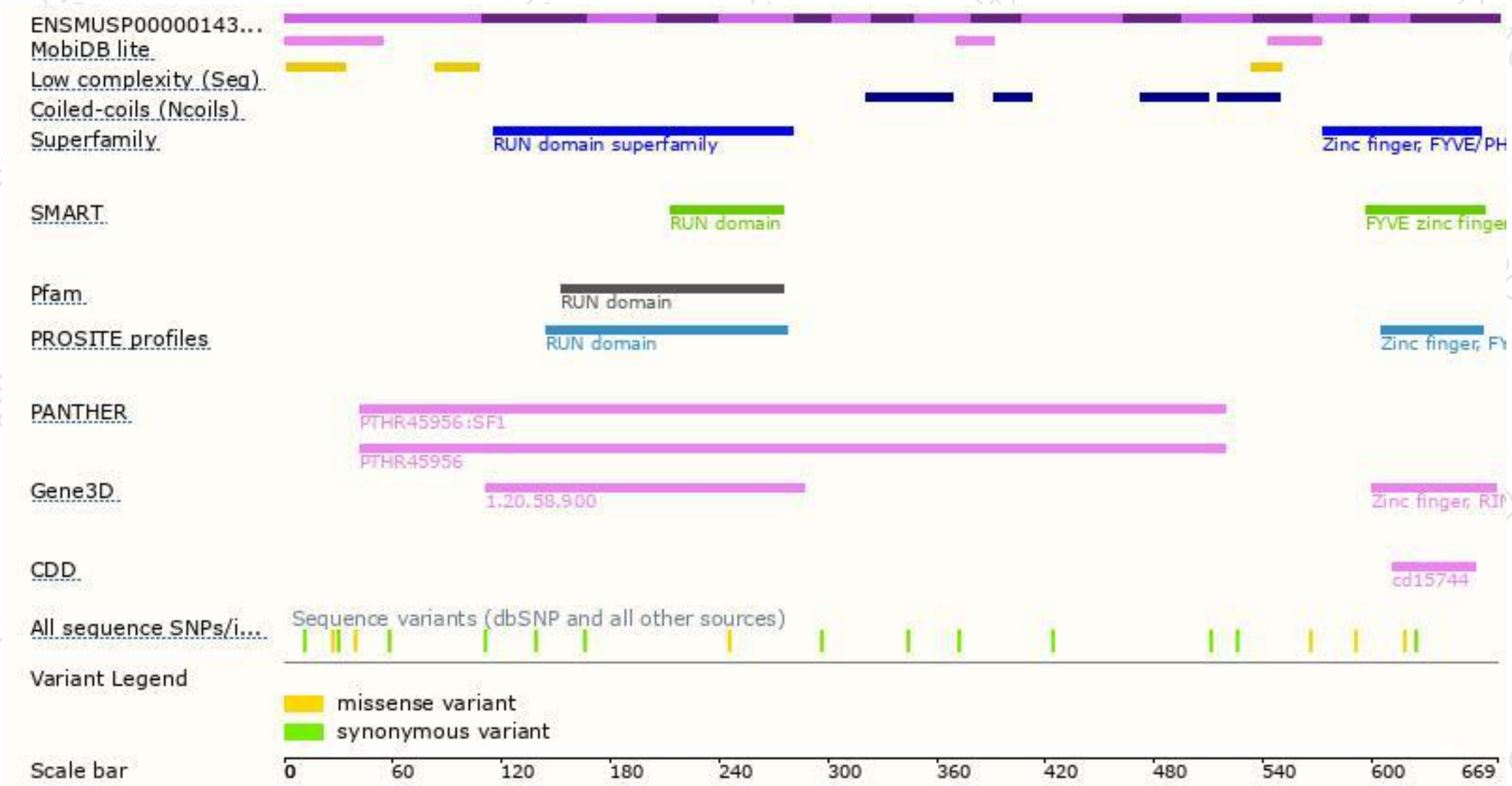The strategy is based on the design of *Rufy3-205* transcript,the transcription is shown below:
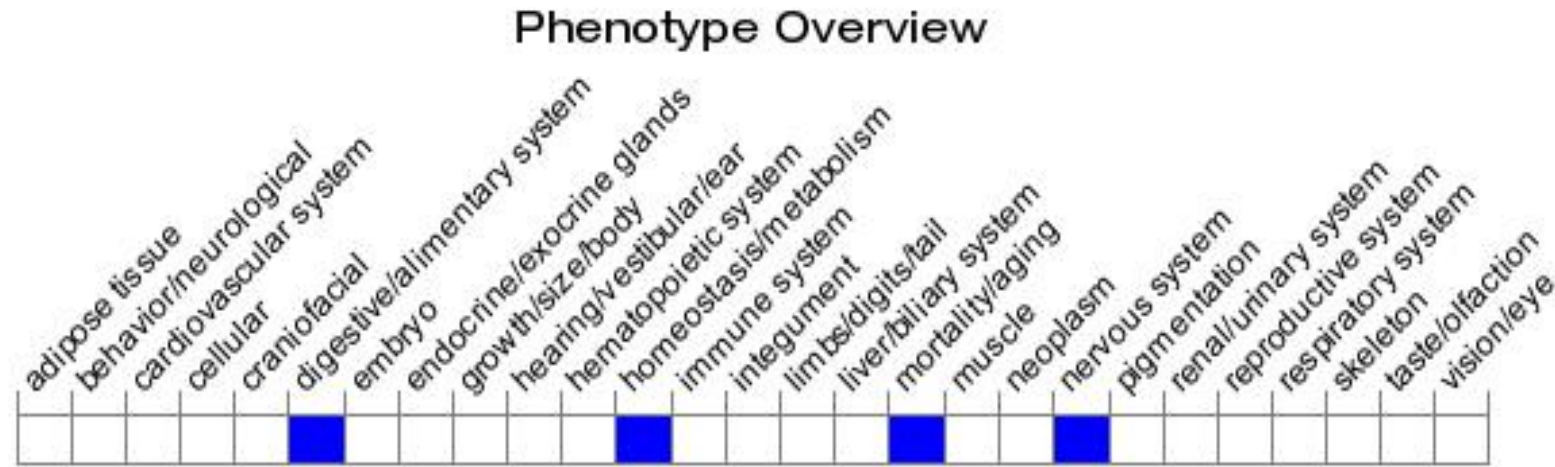
Genomic location distribution
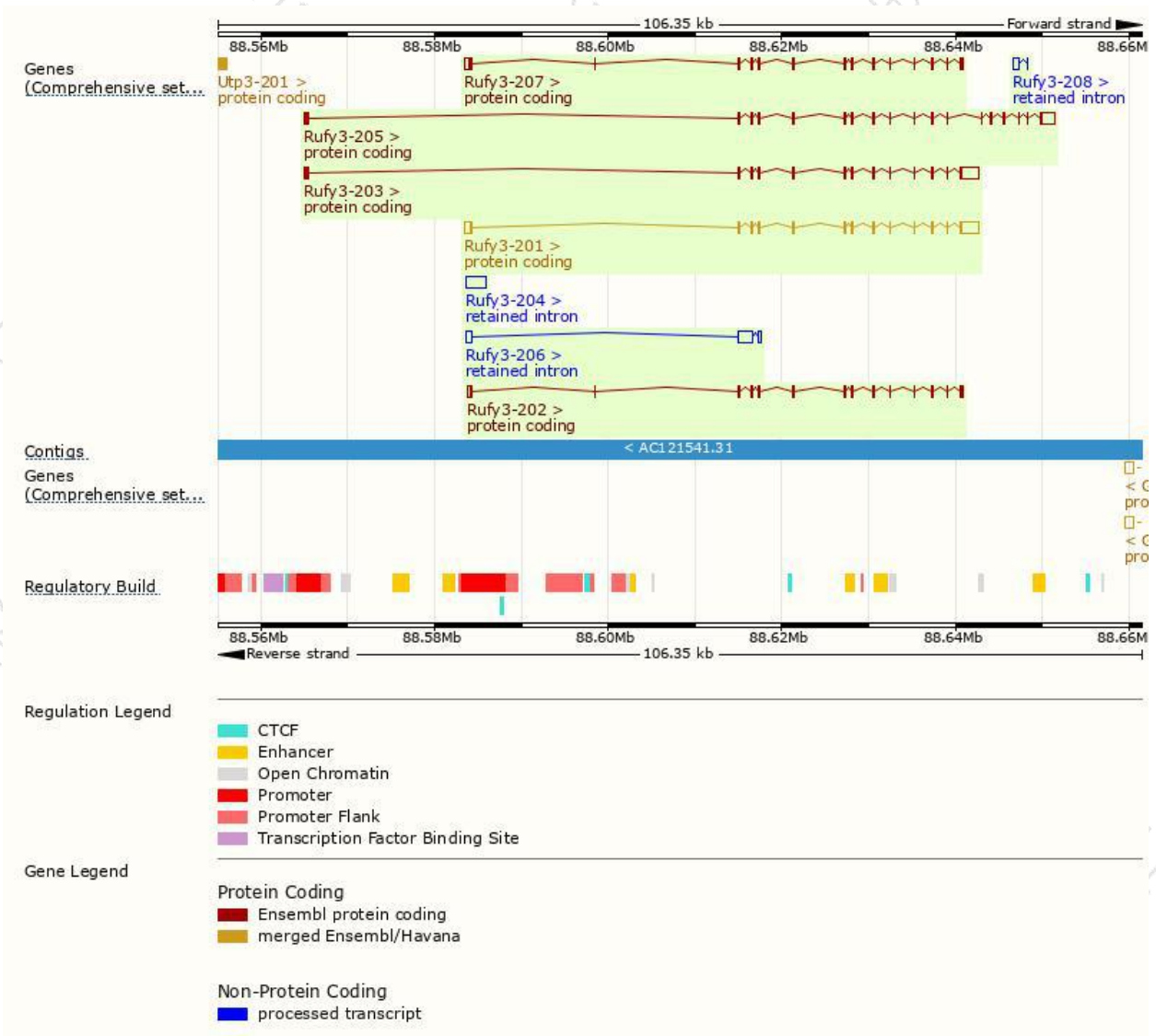
Protein domain

Mouse phenotype description(MGI)

Phenotype Overview

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,the gene product is involved in regulating neuronal polarity and axon growth.

Homozygous KO leads to loss of neuronal polarity and causes neonatal lethality.

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