

Nwd2 Cas9-CKO Strategy

Designer: Zihe Cui

Reviewer: Xiaojing Li

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Project Overview

Project Name

Nwd2

Project type

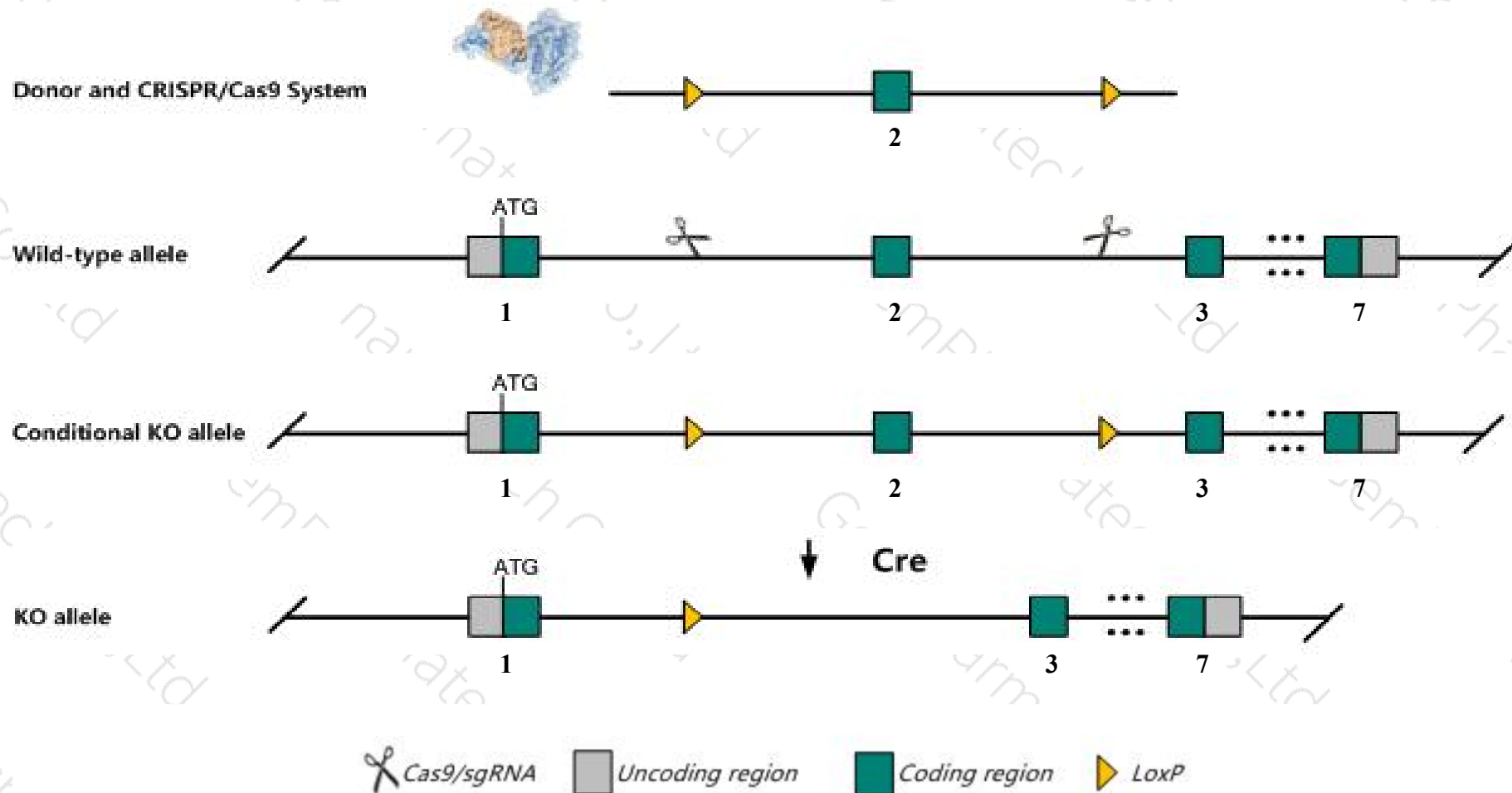
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Nwd2* gene. The schematic diagram is as follows:



Technical routes

- The *Nwd2* gene has 4 transcripts. According to the structure of *Nwd2* gene, exon2 of *Nwd2-201*(ENSMUST00000159584.2) transcript is recommended as the knockout region. The region contains 89bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Nwd2* 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.

- Transcript *Nwd2*-204 may not be affected.
- The *Nwd2* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Nwd2 NACHT and WD repeat domain containing 2 [*Mus musculus* (house mouse)]

Gene ID: 319807, updated on 25-Sep-2020

Summary

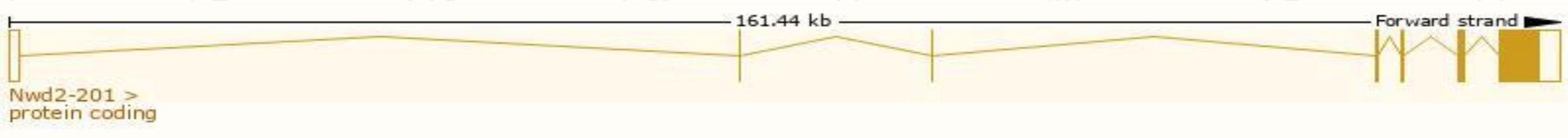
Official Symbol	Nwd2 provided by MGI
Official Full Name	NACHT and WD repeat domain containing 2 provided by MGI
Primary source	MGI:MGI:1920464
See related	Ensembl:ENSMUSG00000090061
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Hn1-ps2; mKIAA1239; 3110047P20Rik; B830017A01Rik
Expression	Biased expression in cortex adult (RPKM 2.5), frontal lobe adult (RPKM 2.4) and 6 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

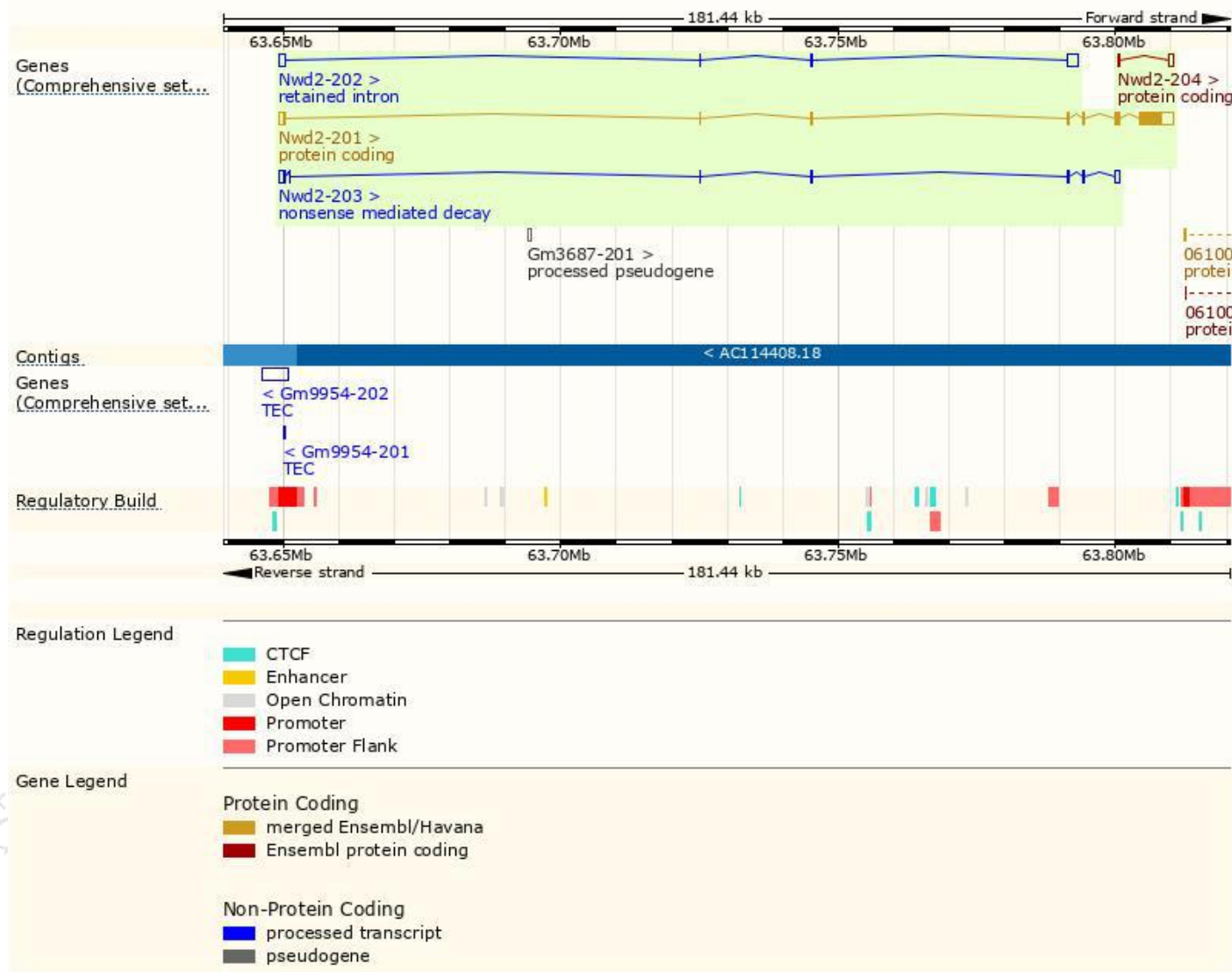
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nwd2-201	ENSMUST00000159584.2	8534	1742aa	Protein coding	CCDS51508	Q6P5U7	TSL:1 GENCODE basic APPRIS P1
Nwd2-204	ENSMUST00000162757.3	1230	69aa	Protein coding	-	A0A0G2JGP2	CDS 5' incomplete TSL:3
Nwd2-203	ENSMUST00000162166.2	2281	53aa	Nonsense mediated decay	-	E0CY00	TSL:1
Nwd2-202	ENSMUST00000160066.7	3369	No protein	Retained intron	-	-	TSL:1

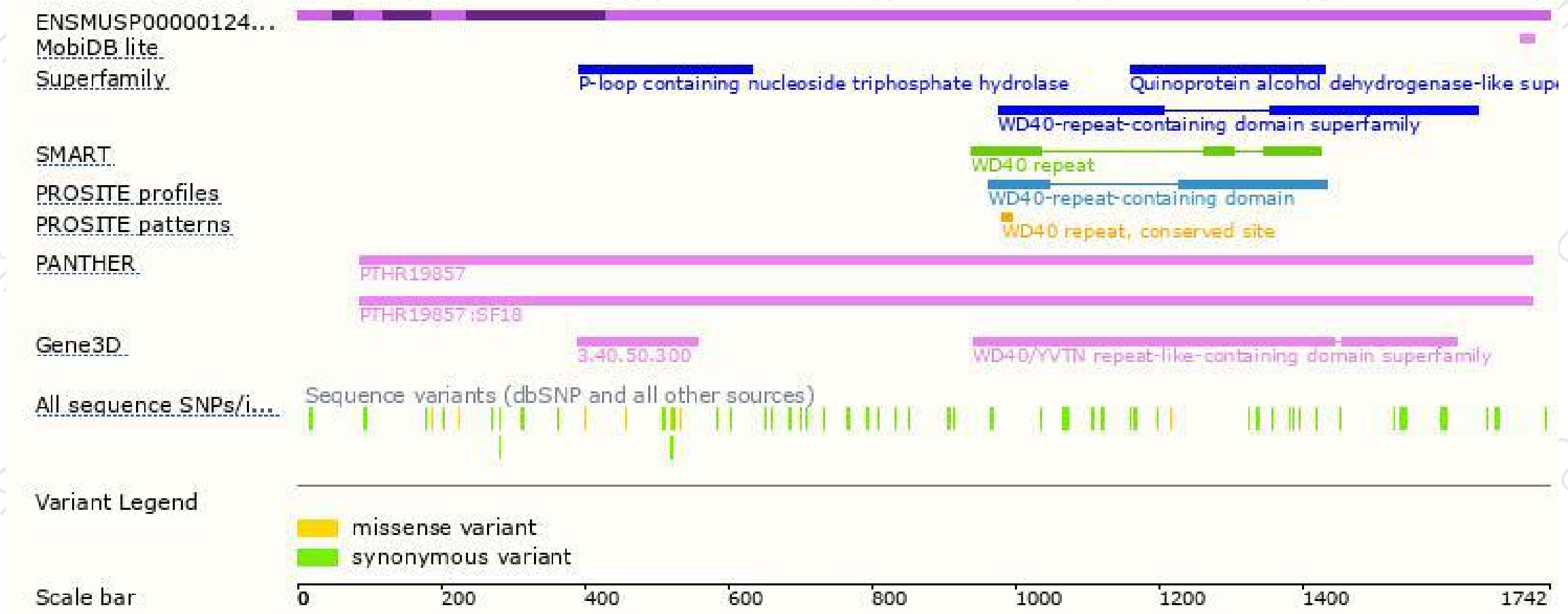
The strategy is based on the design of *Nwd2-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

Tel: 025-5864 1534

