Nxn Cas9-CKO Strategy

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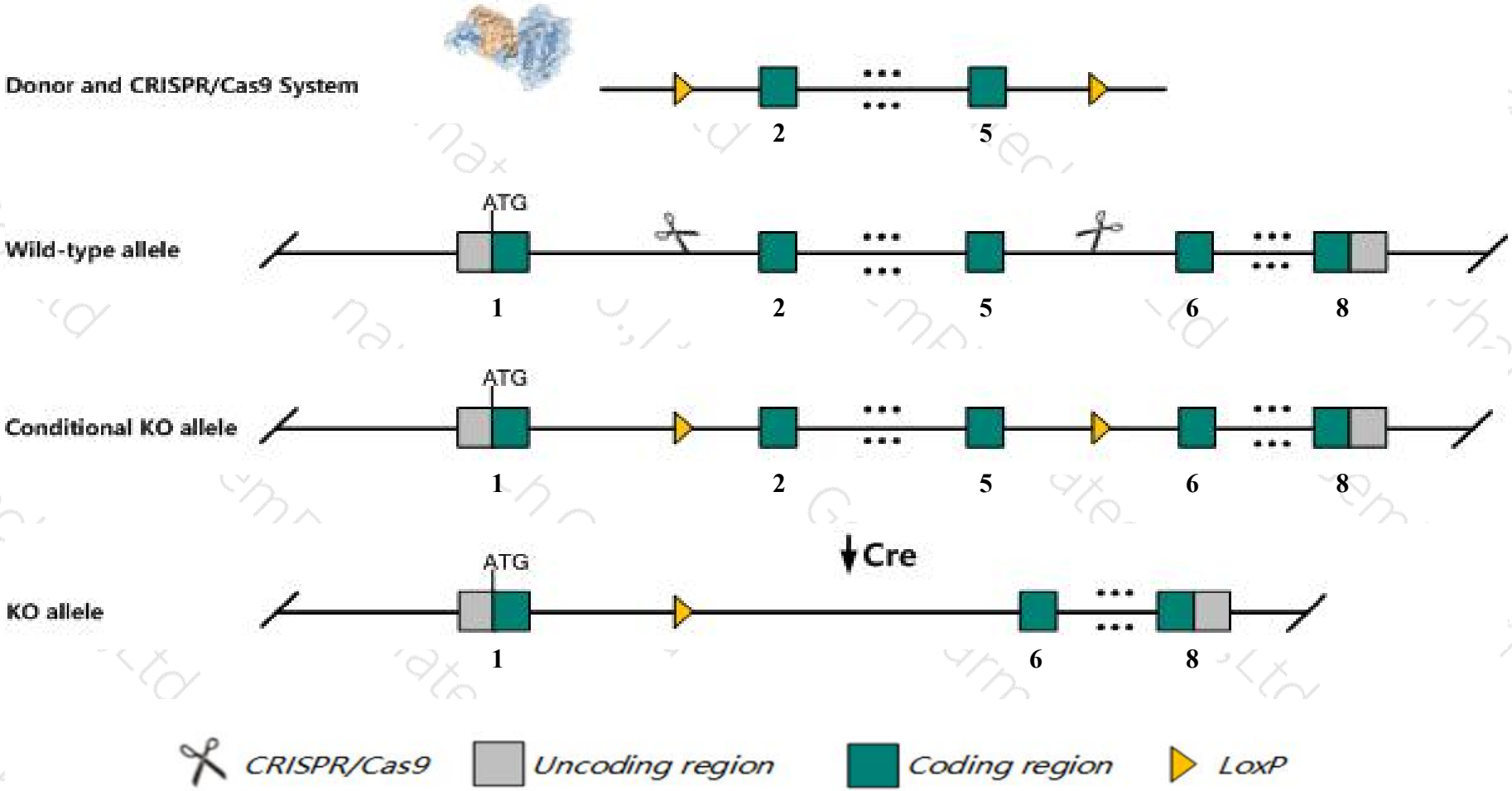
Project Overview

Project Name	***Nxn***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

- The *Nxn* gene has 2 transcripts. According to the structure of *Nxn* gene, exon2-exon5 of *Nxn-201* (ENSMUST00000021204.3) transcript is recommended as the knockout region.The region contains 460bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Nxn* gene. The brief process is as follows:CRISPR/Cas9 system 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 will be 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,homozygous null mice die by p1 and exhibit craniofacial bone defects and cleft palate.
- The *Nxn* gene is located on the Chr11. 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）

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Nxn nucleoredoxin [Mus musculus (house mouse)]

Gene ID: 18230, updated on 13-Mar-2020

Summary

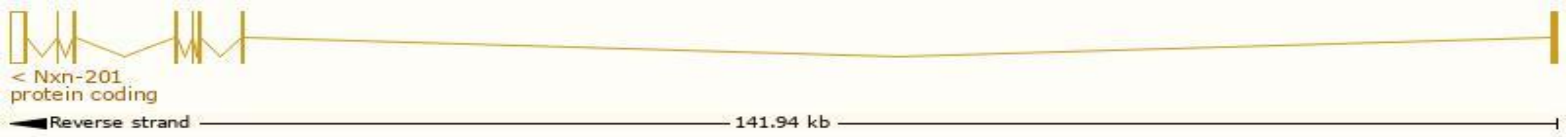
Official Symbol	Nxn provided by MGI
Official Full Name	nucleoredoxin provided byMGI
Primary source	MGI:MGI:109331
See related	Ensembl:ENSMUSG00000020844
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	I11Jus13
Expression	Broad expression in testis adult (RPKM 33.3), limb E14.5 (RPKM 23.7) and 24 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

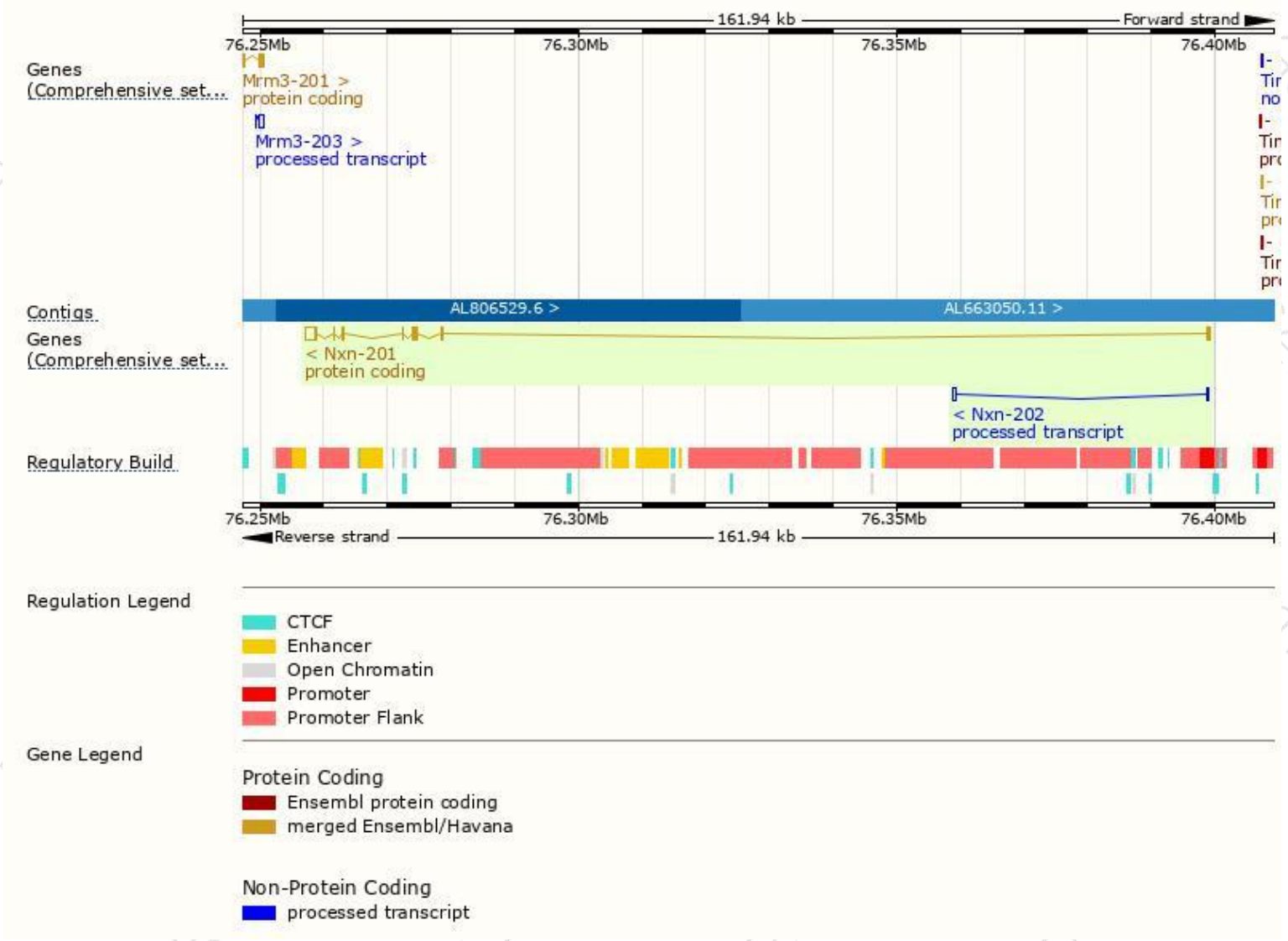
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nxn-201	ENSMUST00000021204.3	2755	435aa	Protein coding	CCDS25064	P97346	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
Nxn-202	ENSMUST00000131472.1	852	No protein	Processed transcript	-	-	TSL:1

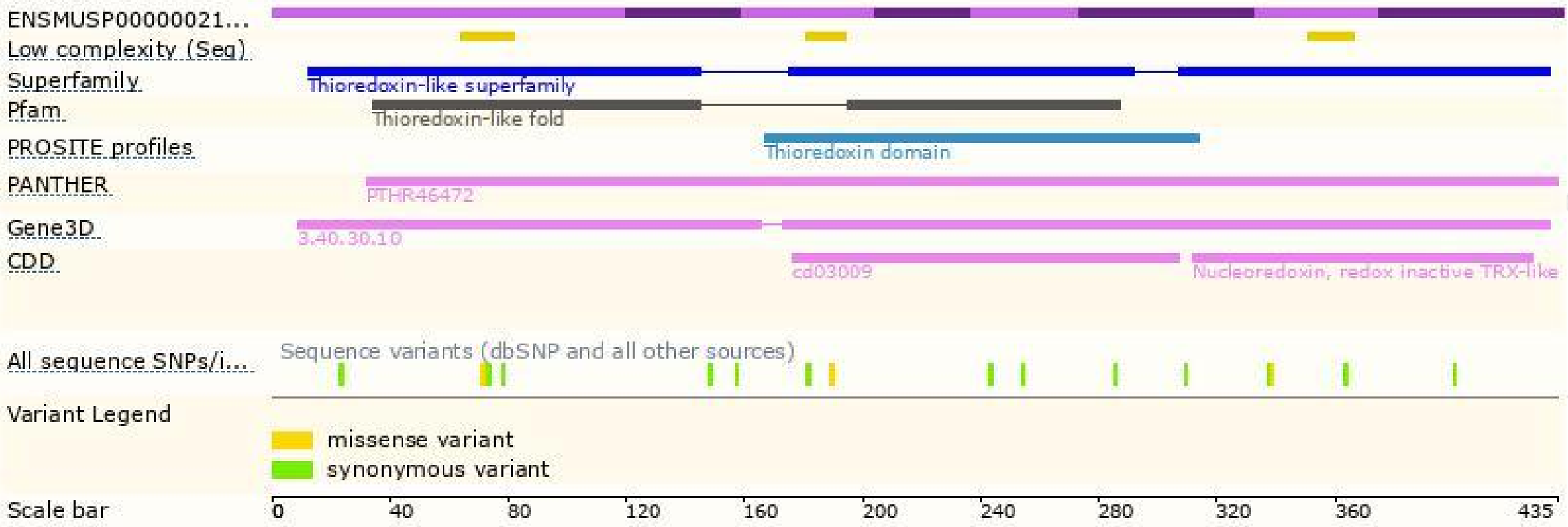
The strategy is based on the design of *Nxn-201* transcript,the transcription is shown below:



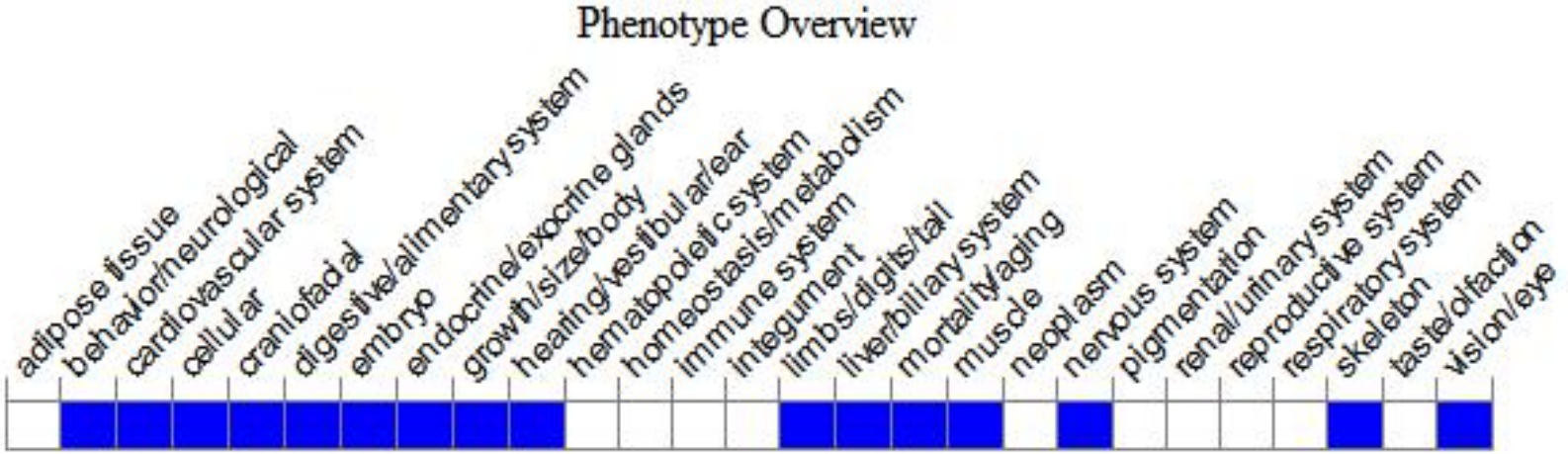
Genomic location distribution



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,homozygous null mice die by P1 and exhibit craniofacial bone defects and cleft palate.

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

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