

# Nup35 Cas9-CKO Strategy

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Design Date: 2024-3-11

# Overview

## Target Gene Name

- Nup35

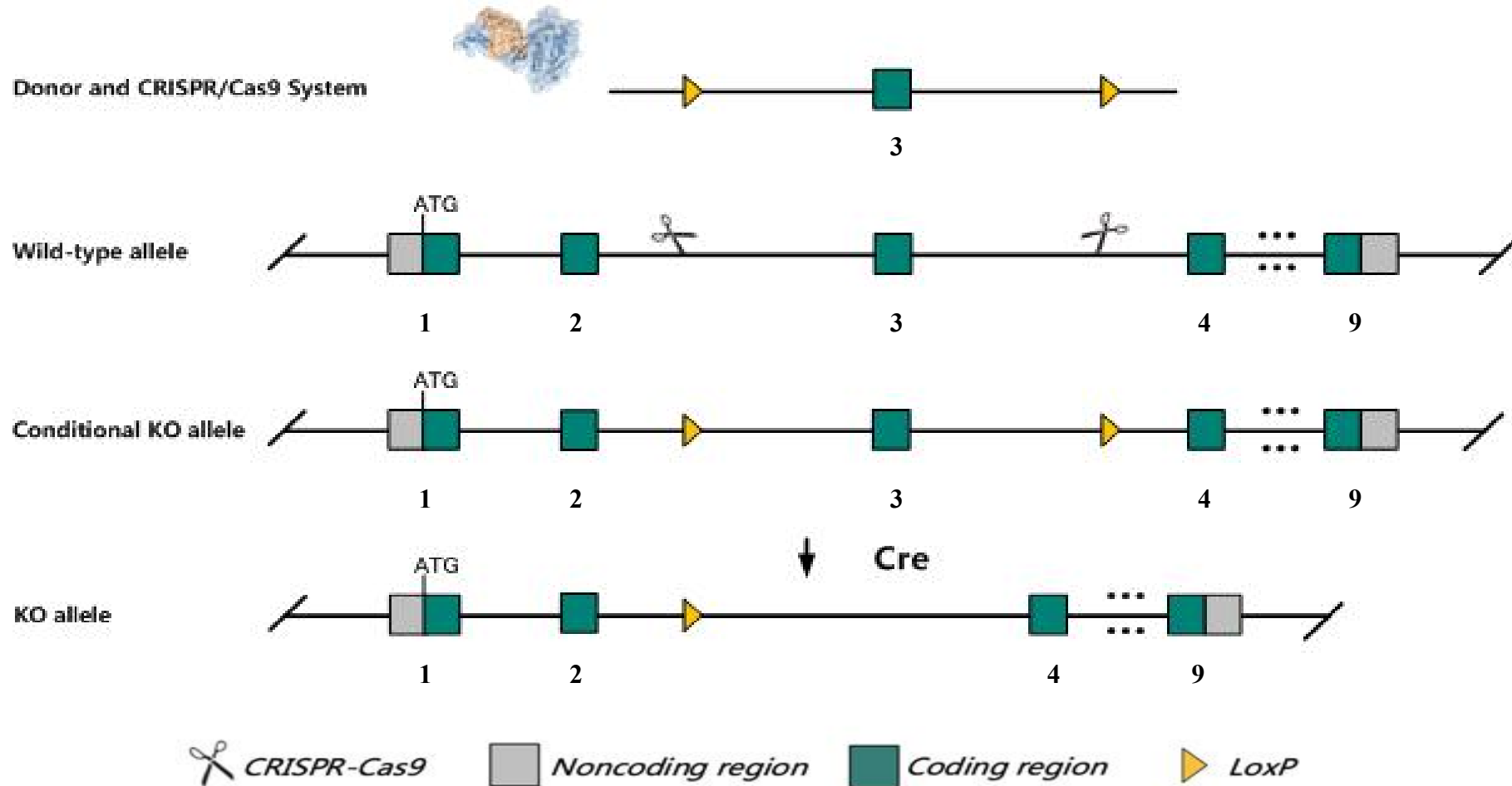
## Project Type

- Cas9-CKO

## Genetic Background

- C57BL/6JGpt

# Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *Nup35* gene.

# Technical Information

- The *Nup35* gene has 5 transcripts. According to the structure of *Nup35* gene, exon3 of *Nup35*-201 (ENSMUST00000028382.13) transcript is recommended as the knockout region. The region contains 128bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Nup35* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.

# Gene Information

**Nup35** nucleoporin 35 [ *Mus musculus* (house mouse) ]

[Download Datasets](#)

Gene ID: 69482, updated on 5-Mar-2024

## Summary

Official Symbol	Nup35 provided by <a href="#">MGI</a>
Official Full Name	nucleoporin 35 provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:1916732</a>
See related	<a href="#">Ensembl:ENSMUSG00000026999</a> <a href="#">AllianceGenome:MGI:1916732</a>
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	MP44; NO44; 35kDa; 2310006I24Rik; 5330402E05Rik
Summary	Enables identical protein binding activity. Acts upstream of or within cellular response to leukemia inhibitory factor. Predicted to be located in nucleus and plasma membrane. Predicted to be part of nuclear pore central transport channel and nuclear pore nuclear basket. Is expressed in cerebellum. Used to study intestinal pseudo-obstruction. Orthologous to human NUP35 (nucleoporin 35). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Ubiquitous expression in testis adult (RPKM 15.4), whole brain E14.5 (RPKM 7.3) and 28 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>
<b>NEW</b>	Try the new <a href="#">Gene table</a>
	Try the new <a href="#">Transcript table</a>

Source: <https://www.ncbi.nlm.nih.gov/>

# Transcript Information

The gene has 5 transcripts, all transcripts are shown below:

Transcript ID ▲	Name ▲	bp ▲	Protein ▲	Biotype ▲	CCDS ▲	UniProt Match ▲	Flags ▲
<a href="#">ENSMUST00000028382.13</a>	Nup35-201	1559	<a href="#">325aa</a>	Protein coding	<a href="#">CCDS16179</a>	<a href="#">Q8R4R6</a>	Ensembl Canonical Gencode basic APPRIS P1 TSL:1
<a href="#">ENSMUST00000124377.2</a>	Nup35-202	653	<a href="#">197aa</a>	Protein coding		<a href="#">A2ATJ2</a>	TSL:3 CDS 3' incomplete
<a href="#">ENSMUST00000127926.2</a>	Nup35-203	692	No protein	Protein coding CDS not defined		-	TSL:3
<a href="#">ENSMUST00000135305.8</a>	Nup35-204	2688	No protein	Retained intron		-	TSL:2
<a href="#">ENSMUST00000144697.2</a>	Nup35-205	496	No protein	Protein coding CDS not defined		-	TSL:3

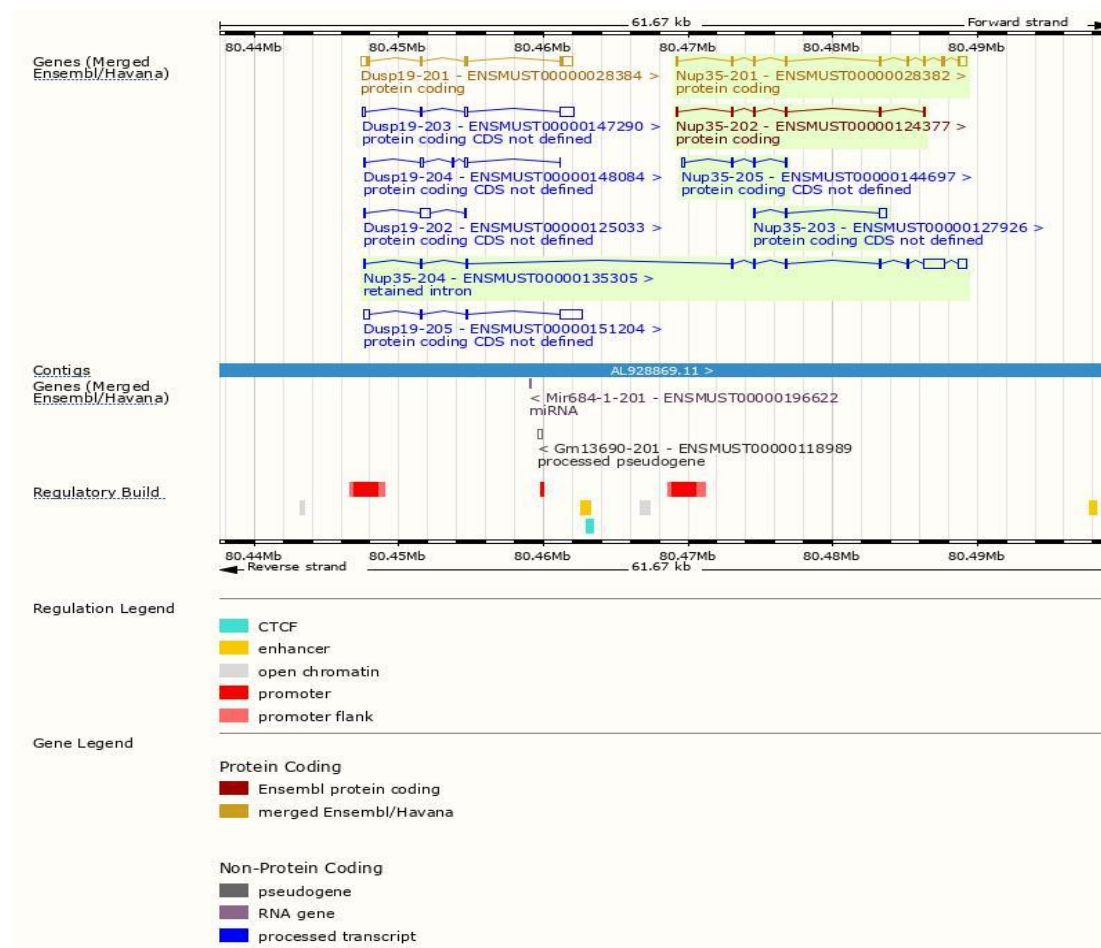
The strategy is based on the design of *Nup35-201* transcript, the transcription is shown below:



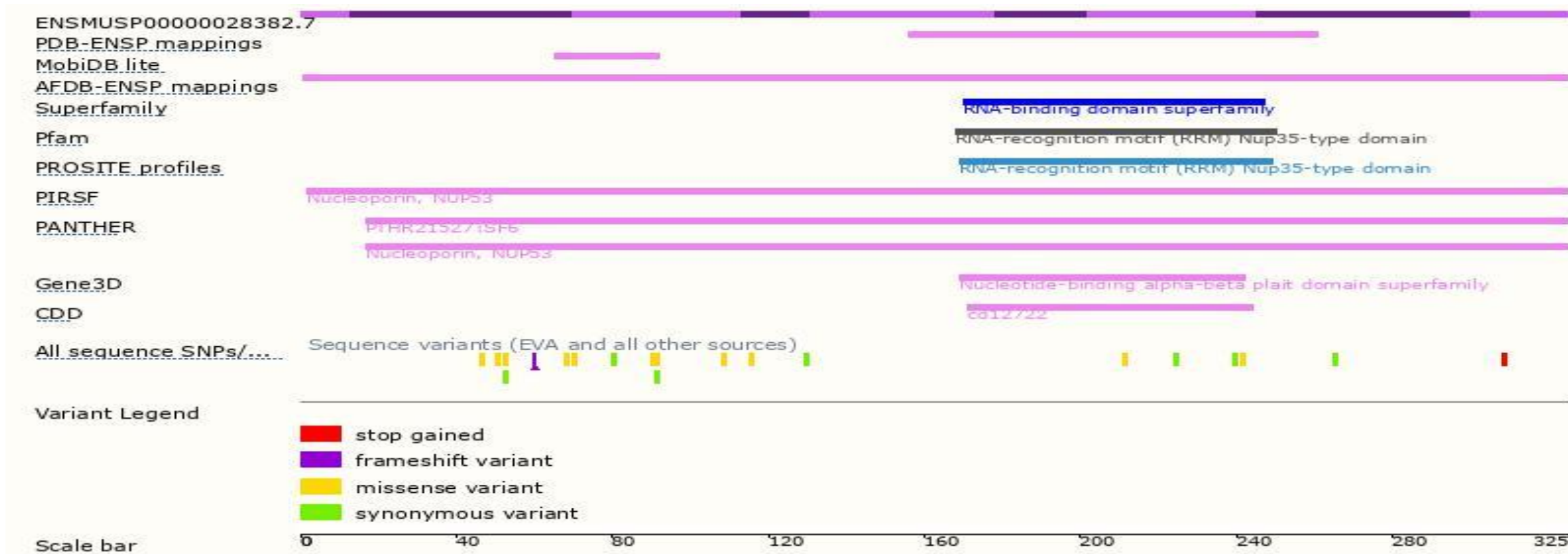
Source: <https://www.ensembl.org>



# Genomic Information

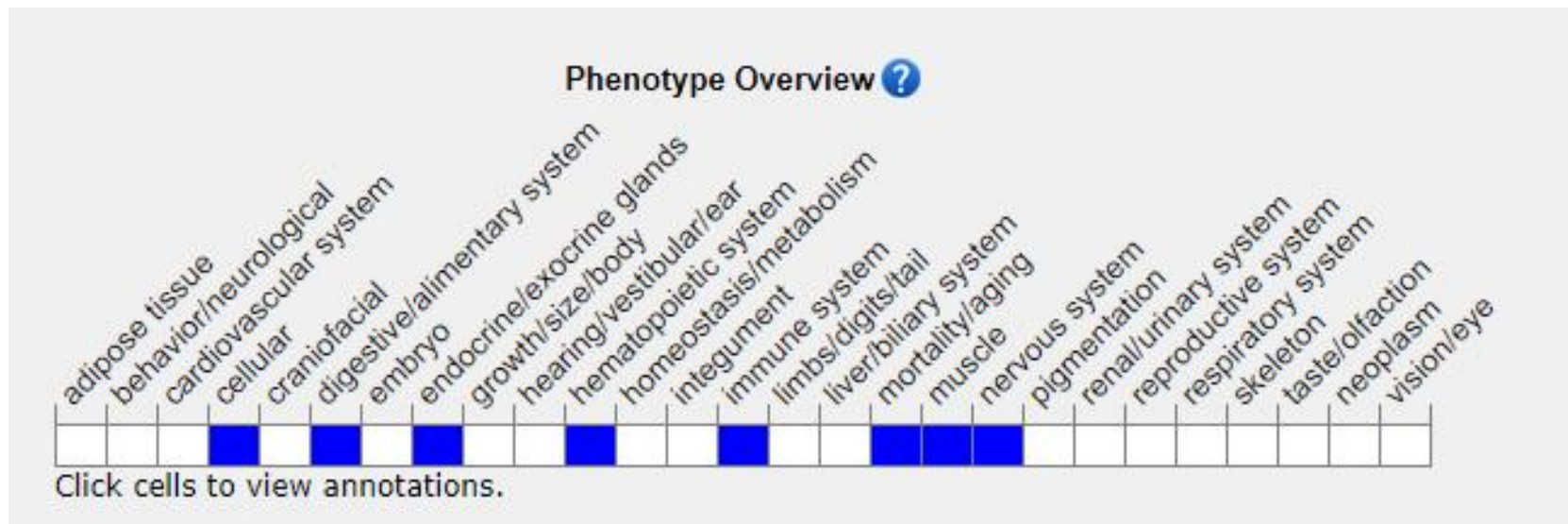


# Protein Information





# Mouse Phenotype Information (MGI)



- Homozygous knockout through a point mutation in a critical functional domain leads to early death as a result of megacolon caused by colon myopathy.

# Important Information

- *Nup35* is located on Chr2. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.

# References

Targeted (Conditional ready, Null/knockout, Reporter)

Insertion Vector: L1L2\_Bact\_P

- ▼ Mutation details: The L1L2\_Bact\_P cassette was inserted at position 80473824 of Chromosome 2 upstream of the critical exon(s) (Build GRCm39). The cassette is composed of an FRT site followed by lacZ sequence and a loxP site. This first loxP site is followed by a neomycin resistance gene under the control of the human beta-actin promoter, SV40 polyA, a second FRT site and a second loxP site. A third loxP site is inserted downstream of the targeted exon(s) at position 80474761. The critical exon(s) is/are thus flanked by loxP sites. A "conditional ready" (floxed) allele can be created by flp recombinase expression in mice carrying this allele. Subsequent cre expression results in a knockout mouse. If cre expression occurs without flp expression, a reporter knockout mouse will be created. Further information on targeting strategies used for this and other IKMC alleles can be found at [http://www.informatics.jax.org/mgihome/nomen/IKMC\\_schematics.shtml](http://www.informatics.jax.org/mgihome/nomen/IKMC_schematics.shtml) (J:155845, J:173534)

<https://www.informatics.jax.org/allele/MGI:5008019>