

Nucb1 Cas9-CKO Strategy

Designer: Huan Wang

Reviewer: Yumeng Wang

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Project Overview

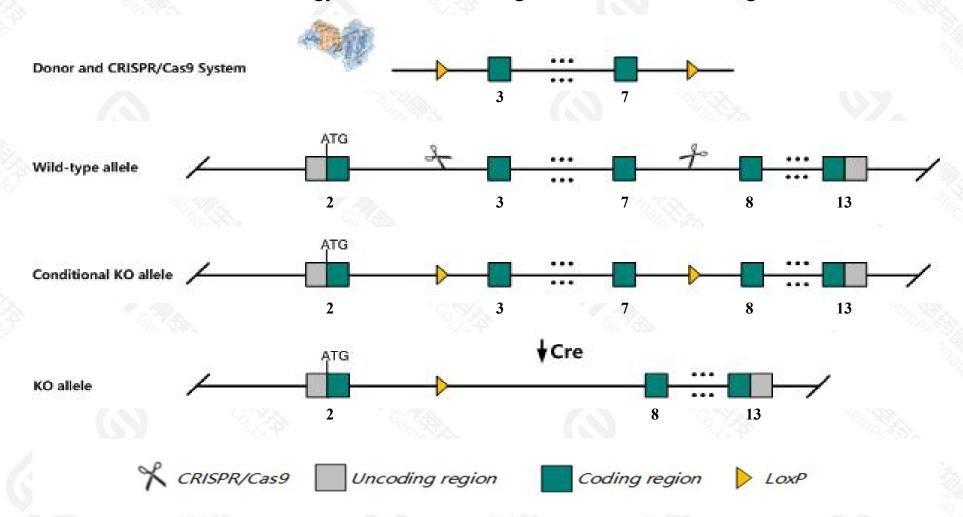


Project Name	Nucb1
Project type	Cas9-CKO
Strain background	C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Nucb1* gene. The schematic diagram is as follows:



Technical routes



- The *Nucb1* gene has 10 transcripts. According to the structure of *Nucb1* gene, exon3-exon7 of *Nucb1-210*(ENSMUST00000211765.2) transcript is recommended as the knockout region. The region contains 622bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Nucb1* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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



- > The *Nucb1* gene is located on the Chr7. 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.
- \rightarrow The KO region contains the intron of *Tulp2* gene. Knockout the region may affect the function of *Tulp2* gene.
- > 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)



Nucb1 nucleobindin 1 [Mus musculus (house mouse)]

Gene ID: 18220, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Nucb1 provided by MGI

Official Full Name nucleobindin 1 provided by MGI

Primary source MGI:MGI:97388

See related Ensembl: ENSMUSG00000030824

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 B230337F23Rik, C77483, Calnuc, MTEST82, Nucb

Expression Ubiquitous expression in colon adult (RPKM 160.1), genital fat pad adult (RPKM 128.0) and 28 other tissuesSee more

Orthologs human all

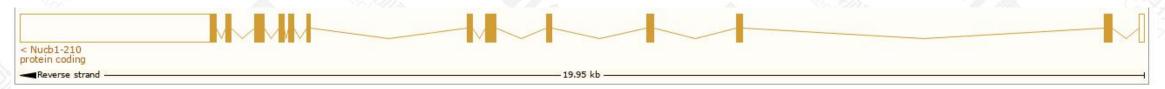
Transcript information (Ensembl)



The gene has 10 transcripts, all transcripts are shown below:

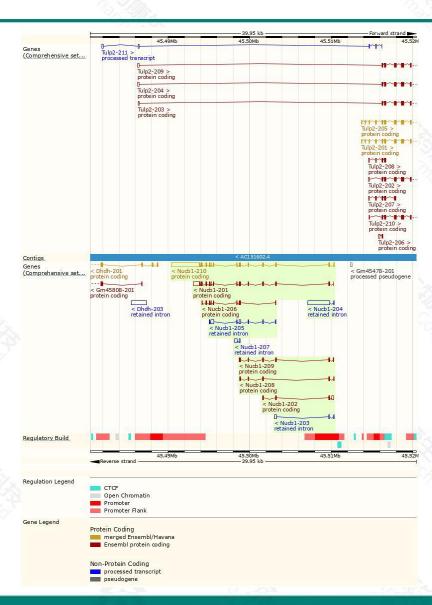
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nucb1-210	ENSMUST00000211765.1	4853	459aa	Protein coding	CCDS52247	Q02819	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Nucb1-201	ENSMUST00000033096.15	2111	455aa	Protein coding	CCDS85300	A0A1C7CYU3	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 ALT2
Nucb1-206	ENSMUST00000210449.1	811	271aa	Protein coding	(2)	A0A1B0GR41	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5 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 ALT:
Nucb1-209	ENSMUST00000211682.1	755	220aa	Protein coding	-	A0A1B0GR92	CDS 3' incomplete TSL:2
Nucb1-202	ENSMUST00000209436.1	615	<u>121aa</u>	Protein coding	170	A0A1B0GT83	CDS 3' incomplete TSL:2
Nucb1-208	ENSMUST00000211343.1	559	170aa	Protein coding		A0A1B0GS16	CDS 3" incomplete TSL:5
Nucb1-204	ENSMUST00000210208.1	2742	No protein	Retained intron	0-0	14	TSL:1
Nucb1-205	ENSMUST00000210394.1	1069	No protein	Retained intron	-	-	TSL:5
Nucb1-207	ENSMUST00000210675.1	438	No protein	Retained intron	-	-	TSL:3
Nucb1-203	ENSMUST00000209509.1	433	No protein	Retained intron	-	-	TSL2

The strategy is based on the design of *Nucb1-210* transcript, the transcription is shown below:



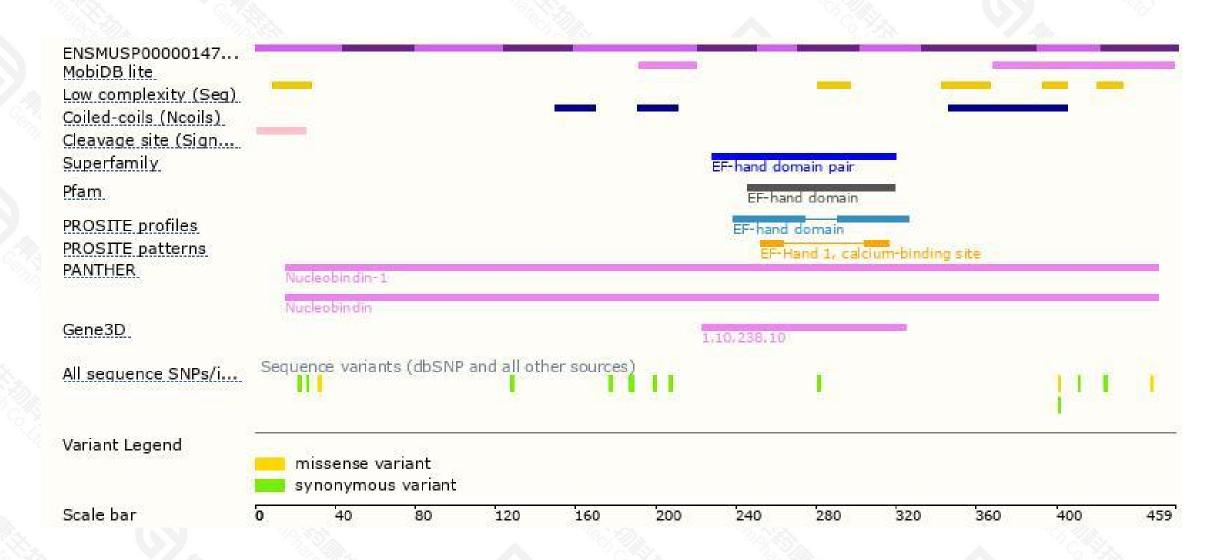
Genomic location distribution





Protein domain







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

Tel: 400-9660890





