

Cntfr Cas9-CKO Strategy

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Project Overview



Project Name Cntfr

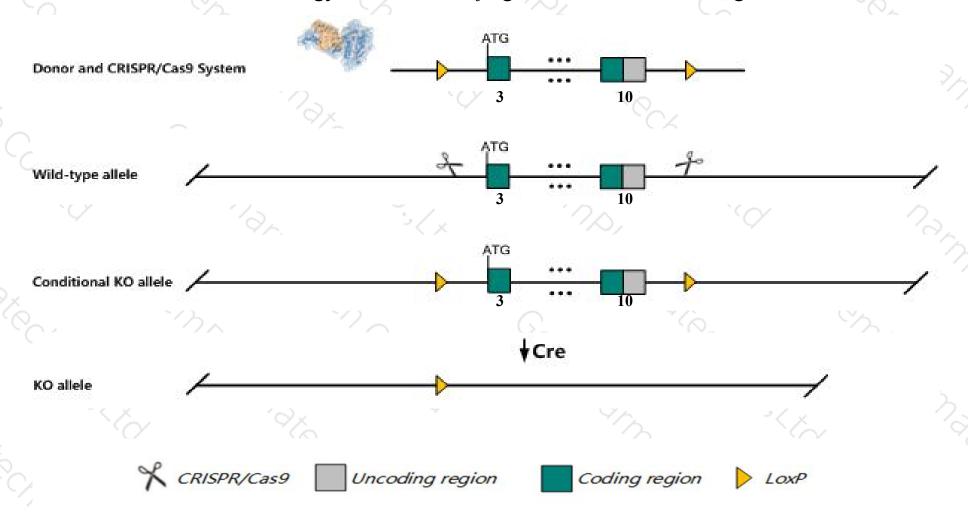
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



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



Technical routes



- The *Cntfr* gene has 6 transcripts. According to the structure of *Cntfr* gene, exon3-exon10 of *Cntfr-203* (ENSMUST00000102961.9) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cntfr* 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 mutant animals exhibit a significant reduction in the number of motor neurons. Neonatal mutants fail to suckle and die within 24 hours after birth.
- > The *Cntfr* gene is located on the Chr4. 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)



Cntfr ciliary neurotrophic factor receptor [Mus musculus (house mouse)]

Gene ID: 12804, updated on 31-Jan-2019

Summary

Official Symbol Cntfr provided by MGI

Official Full Name ciliary neurotrophic factor receptor provided by MGI

MGI:MGI:99605 Primary source

> See related Ensembl:ENSMUSG00000028444

Gene type protein coding RefSeg status REVIEWED Organism

Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as Cntfralpha

> This gene encodes the alpha subunit of the ciliary neurotrophic factor (CNTF) receptor that triggers the assembly of a trimolecular complex Summary

upon binding to CNTF, and initiate a downstream signaling process. The encoded preproprotein undergoes proteolytic processing to generate a glycosylphosphatidylinositol-linked cell surface protein. Mice lacking the encoded protein die shortly after birth and exhibit a reduction of motoneuron number at birth. The transgenic disruption of this gene specifically in the skeletal muscle followed by a peripheral nerve lesion impairs motor neuron axonal regeneration across the lesion site. Alternative splicing of this gene results in multiple transcript

variants. [provided by RefSeq, Nov 2015]

Expression Broad expression in adrenal adult (RPKM 8.1), cerebellum adult (RPKM 6.8) and 17 other tissuesSee more

Orthologs human all

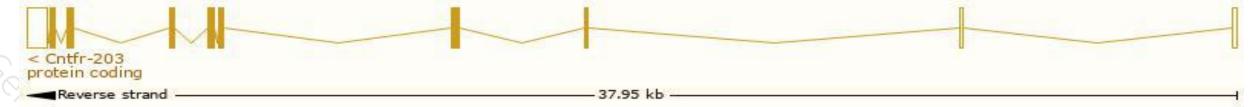
Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

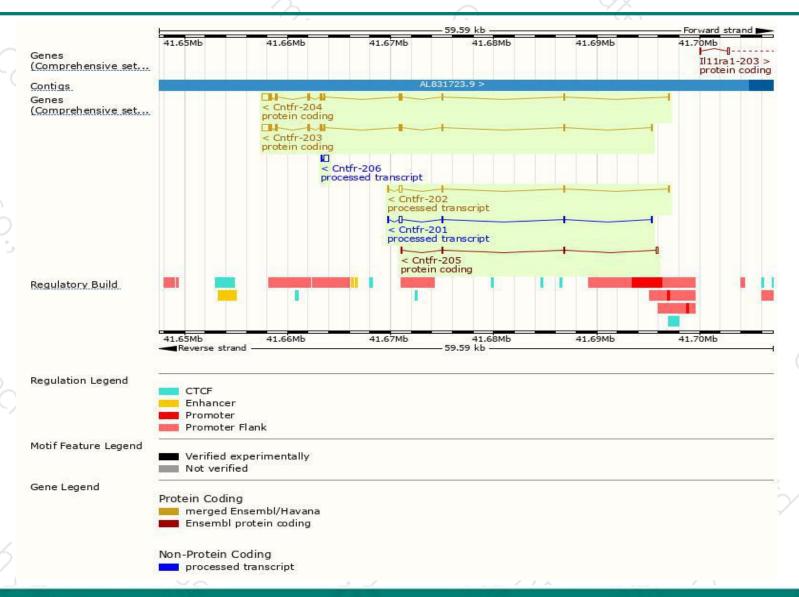
Name 🍦	Transcript ID ▼	bp 🍦	Protein 🍦	Biotype	CCDS 🍦	UniProt 4	Flags
Cntfr-206	ENSMUST00000151181.1	596	No protein	IncRNA	19	-	TSL:1
Cntfr-205	ENSMUST00000145379.1	410	60aa	Protein coding	N9	A2AMR7 ₽	CDS 3' incomplete TSL:5
Cntfr-204	ENSMUST00000102962.9	1926	372aa	Protein coding	CCDS18067 ₽	<u>088507</u> ₽	TSL:1 GENCODE basic APPRIS P
Cntfr-203	ENSMUST00000102961.9	1995	<u>372aa</u>	Protein coding	CCDS18067 ₽	<u>088507</u> ₽	TSL:1 GENCODE basic APPRIS P
Cntfr-202	ENSMUST00000084701.11	676	No protein	IncRNA	8	18	TSL:1
Cntfr-201	ENSMUST00000064443.12	699	No protein	I IncRNA	8-	-	TSL:3

The strategy is based on the design of *Cntfr-203* transcript, The transcription is shown below



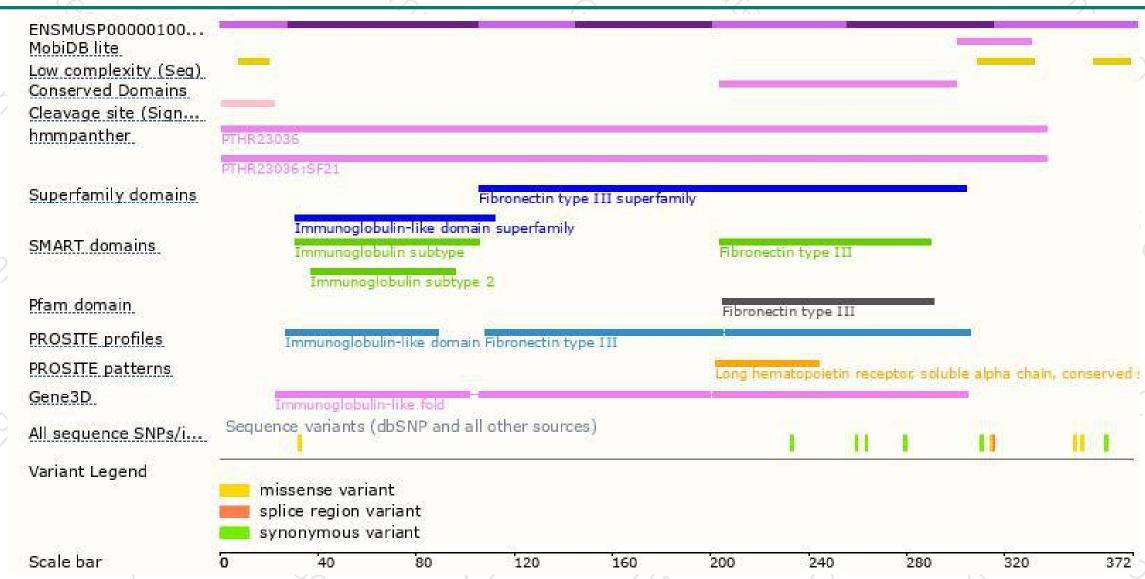
Genomic location distribution





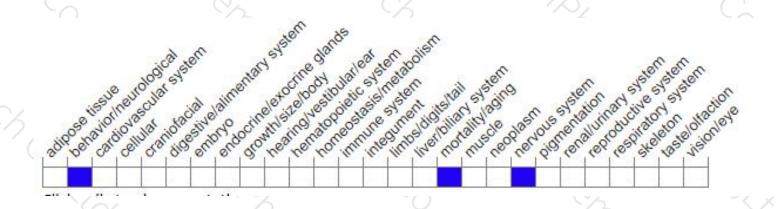
Protein domain





Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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If you have any questions, you are welcome to inquire. Tel: 400-9660890





