

Nfxl1 Cas9-CKO Strategy

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Reviewer: Xiaojing Li

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Project Overview



Project Name Nfxl1

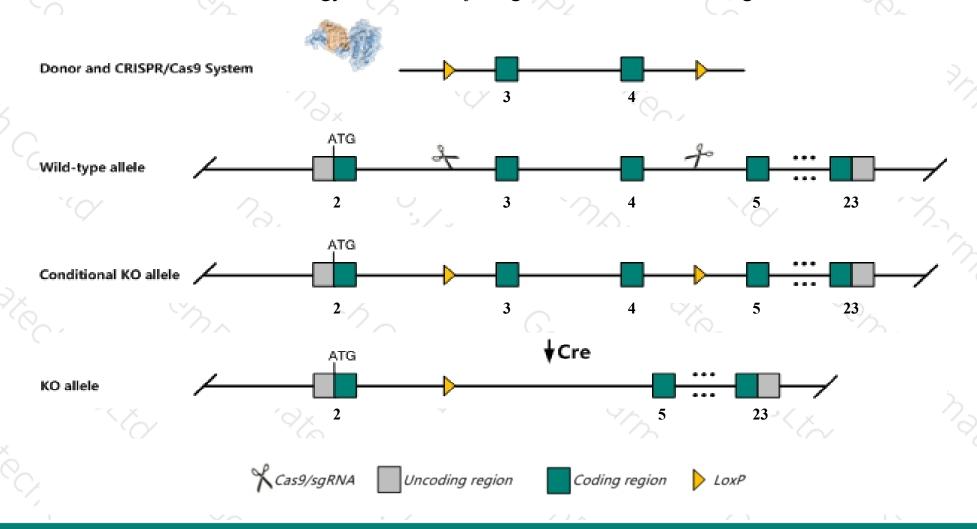
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Nfxl1* gene. The schematic diagram is as follows:



Technical routes



- The *Nfxl1* gene has 9 transcripts. According to the structure of *Nfxl1* gene, exon3-exon4 of *Nfxl1*
 202(ENSMUST00000087216.11) transcript is recommended as the knockout region. The region contains 284bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Nfxl1* 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.

Notice



- > The *Nfxl1* 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)



Nfxl1 nuclear transcription factor, X-box binding-like 1 [Mus musculus (house mouse)]

Gene ID: 100978, updated on 20-Mar-2020

Summary



Official Symbol Nfxl1 provided by MGI

Official Full Name nuclear transcription factor, X-box binding-like 1 provided by MGI

Primary source MGI:MGI:1923646

See related Ensembl:ENSMUSG00000072889

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 1700012H24Rik, AW538212, D430033A06Rik, DNABF, GCF, Gm1058, TCF9

Expression Ubiquitous expression in CNS E11.5 (RPKM 9.0), limb E14.5 (RPKM 7.9) and 28 other tissuesSee more

Orthologs <u>human</u> all

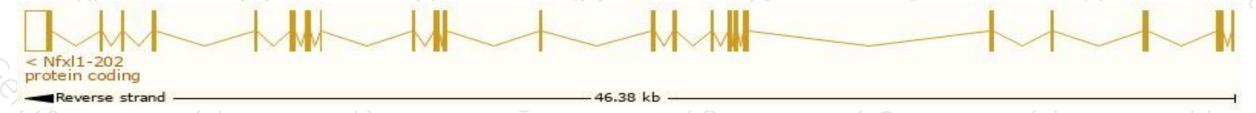
Transcript information (Ensembl)



The gene has 9 transcripts, all transcripts are shown below:

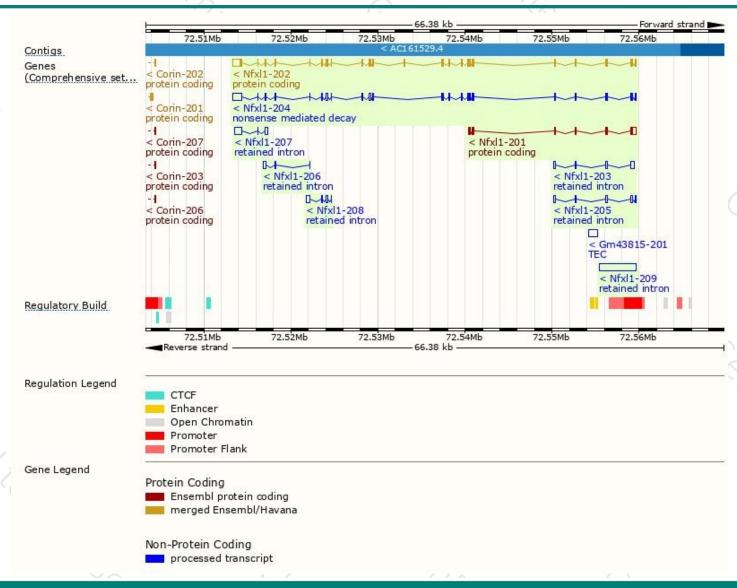
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nfxl1-202	ENSMUST00000087216.11	3712	<u>918aa</u>	Protein coding	CCDS39109	E9Q8I7	TSL:5 GENCODE basic APPRIS P1
Nfxl1-201	ENSMUST00000074948.4	1373	<u>346aa</u>	Protein coding	-	E9Q3K1	CDS 3' incomplete TSL:1
Nfxl1-204	ENSMUST00000135318.7	3579	<u>500aa</u>	Nonsense mediated decay	-	E9PZN0	TSL:1
Nfxl1-209	ENSMUST00000198977.1	4172	No protein	Retained intron	-	-	TSL:NA
Nfxl1-207	ENSMUST00000149393.1	1193	No protein	Retained intron	-	-	TSL:1
Nfxl1-203	ENSMUST00000134452.7	884	No protein	Retained intron	-	-	TSL:1
Nfxl1-208	ENSMUST00000153882.1	800	No protein	Retained intron	-	-	TSL:3
Nfxl1-205	ENSMUST00000146761.1	798	No protein	Retained intron	-	-	TSL:1
Nfxl1-206	ENSMUST00000149340.1	416	No protein	Retained intron	-	-	TSL:3

The strategy is based on the design of *Nfxl1-202* transcript, the transcription is shown below:



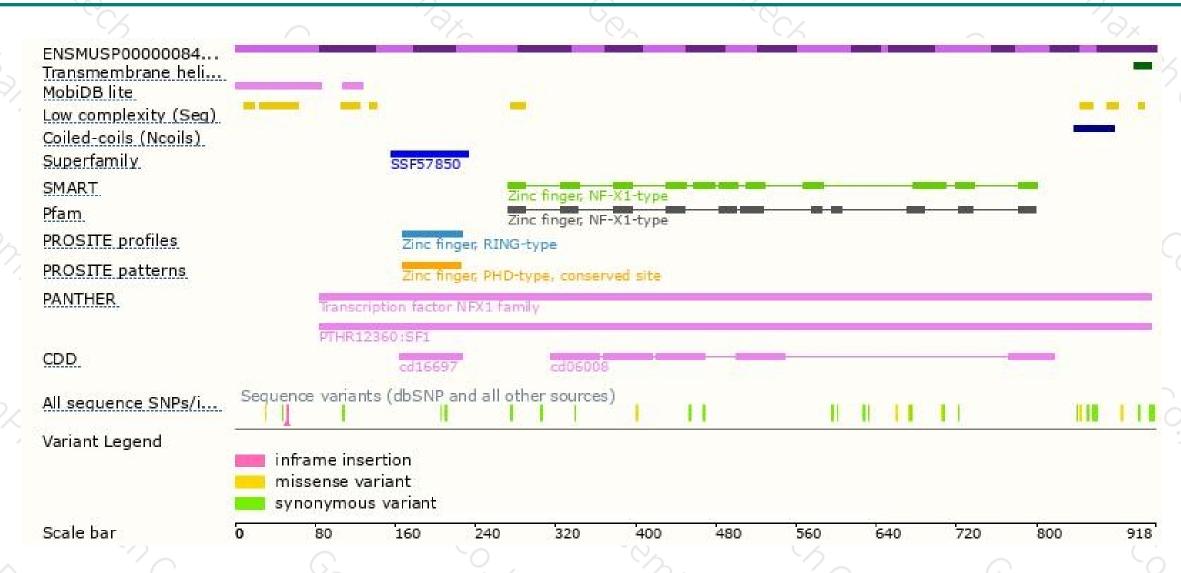
Genomic location distribution





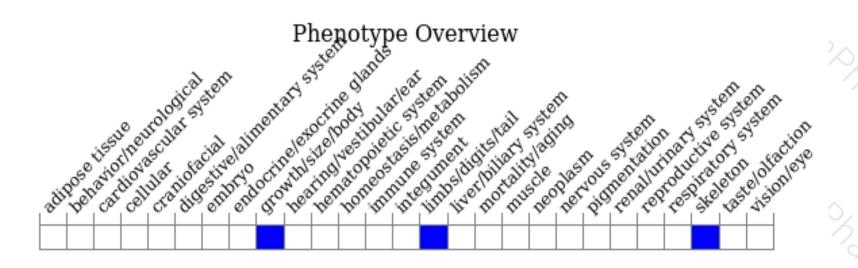
Protein domain





Mouse phenotype description(MGI)





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



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

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