

# Kif1b Cas9-CKO Strategy

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Design Date: 2019-9-11

Reviewer: JiaYu

# **Project Overview**



**Project Name** 

Kif1b

**Project type** 

Cas9-CKO

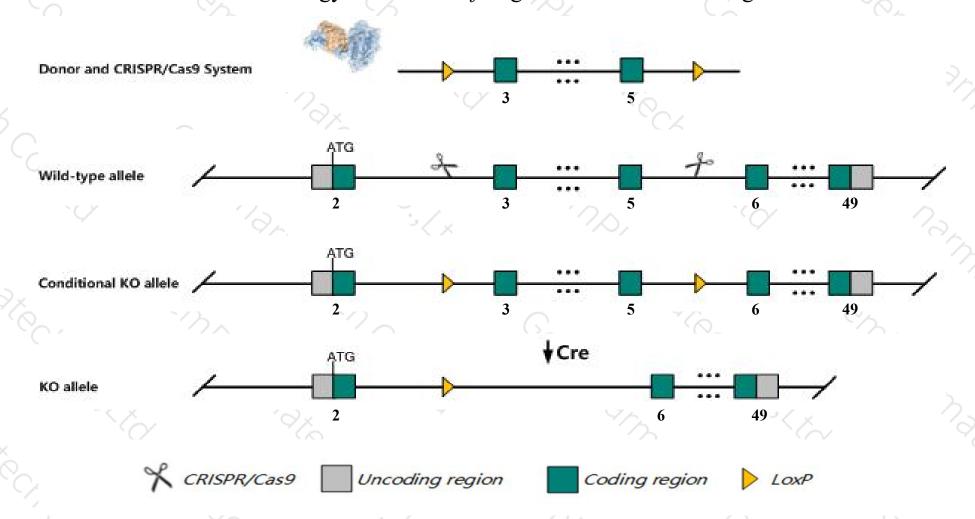
Strain background

C57BL/6JGpt

# Conditional Knockout strategy



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



### Technical routes



- The *Kif1b* gene has 11 transcripts. According to the structure of *Kif1b* gene, exon3-exon5 of *Kif1b-203* (ENSMUST00000060537.12) transcript is recommended as the knockout region. The region contains 323bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Kif1b* 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, Homozygotes for a targeted null mutation exhibit reduced brain size, elevated pain threshold, and neonatal death from apnea. Heterozygotes exhibit impaired synaptic vesicle precursor transport and progressive muscle weakness.
- > The *Kif1b* 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)



#### Kif1b kinesin family member 1B [Mus musculus (house mouse)]

Gene ID: 16561, updated on 31-Jan-2019

#### Summary

☆ ?

Official Symbol Kif1b provided by MGI

Official Full Name kinesin family member 1B provided by MGI

Primary source MGI:MGI:108426

See related Ensembl:ENSMUSG00000063077

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 A530096N05Rik, Al448212, Al506502, D4Mil1e, KIF1Bp130, KIF1Bp204

Expression Broad expression in cerebellum adult (RPKM 37.2), cortex adult (RPKM 32.4) and 25 other tissuesSee more

Orthologs <u>human all</u>

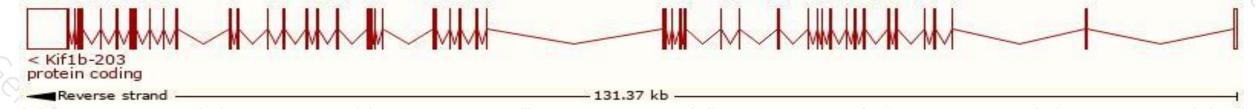
# Transcript information (Ensembl)



The gene has 11 transcripts, all transcripts are shown below:

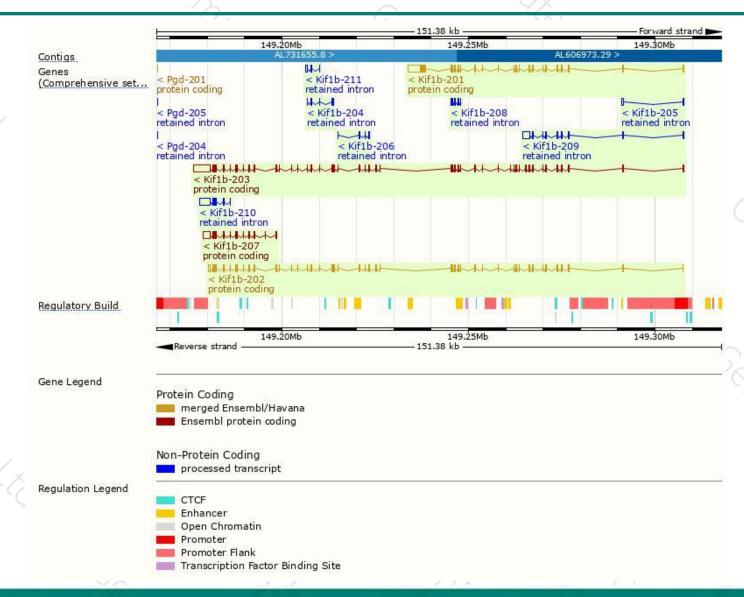
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kif1b-203	ENSMUST00000060537.12	10233	<u>1816aa</u>	Protein coding	CCDS71521	Q60575	TSL:1 GENCODE basic APPRIS ALT1
Kif1b-201	ENSMUST00000030806.5	6991	<u>1150aa</u>	Protein coding	CCDS18957	A2AH75	TSL:1 GENCODE basic
Kif1b-202	ENSMUST00000055647.14	5992	<u>1770aa</u>	Protein coding	CCDS18956	Q60575	TSL:1 GENCODE basic APPRIS P3
Kif1b-207	ENSMUST00000139123.7	3391	<u>495aa</u>	Protein coding	70	F6XNB6	CDS 5' incomplete TSL:1
Kif1b-210	ENSMUST00000150853.1	3475	No protein	Retained intron	-	(5)	TSL:1
Kif1b-209	ENSMUST00000150230.1	2980	No protein	Retained intron	-	680	TSL:1
Kif1b-206	ENSMUST00000133526.1	730	No protein	Retained intron		(4)	TSL:3
Kif1b-211	ENSMUST00000154411.7	620	No protein	Retained intron	92	3. <u>2</u> 3	TSL:3
Kif1b-208	ENSMUST00000140749.1	602	No protein	Retained intron	-	150	TSL:2
Kif1b-205	ENSMUST00000132072.1	587	No protein	Retained intron	-	680	TSL:2
Kif1b-204	ENSMUST00000123593.1	586	No protein	Retained intron	-	1920	TSL:3
	* / * /				1		

The strategy is based on the design of *Kif1b-203* transcript, The transcription is shown below



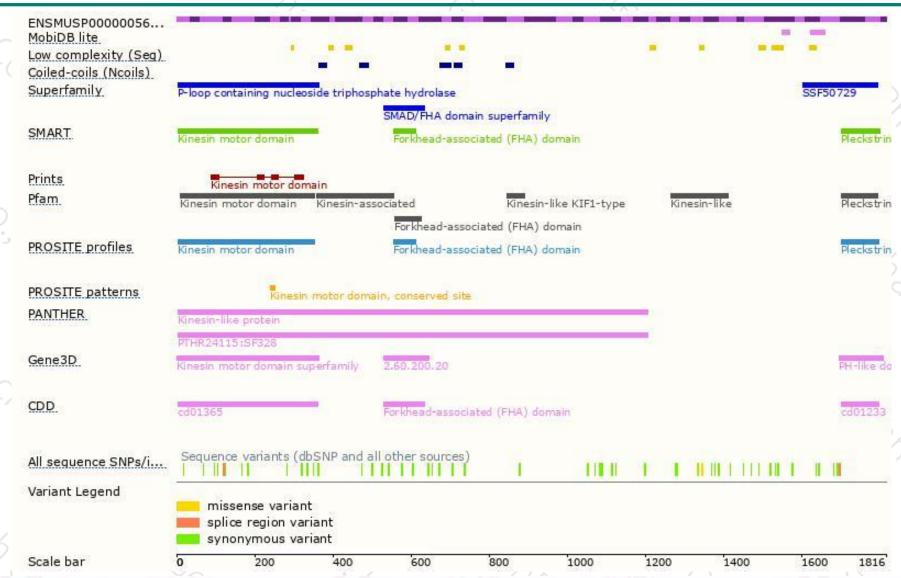
### Genomic location distribution





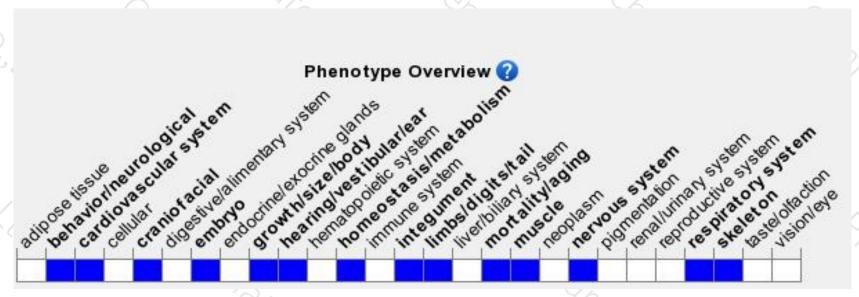
### Protein domain





# Mouse phenotype description(MGI)





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

According to the existing MGI data, Homozygotes for a targeted null mutation exhibit reduced brain size, elevated pain threshold, and neonatal death from apnea. Heterozygotes exhibit impaired synaptic vesicle precursor transport and progressive muscle weakness.



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





