

***Kif22* Cas9-CKO Strategy**

Designer: Yun Li

Reviewer: Longyun Hu

Design Date: 2020-11-10

Project Overview

Project Name

Kif22

Project type

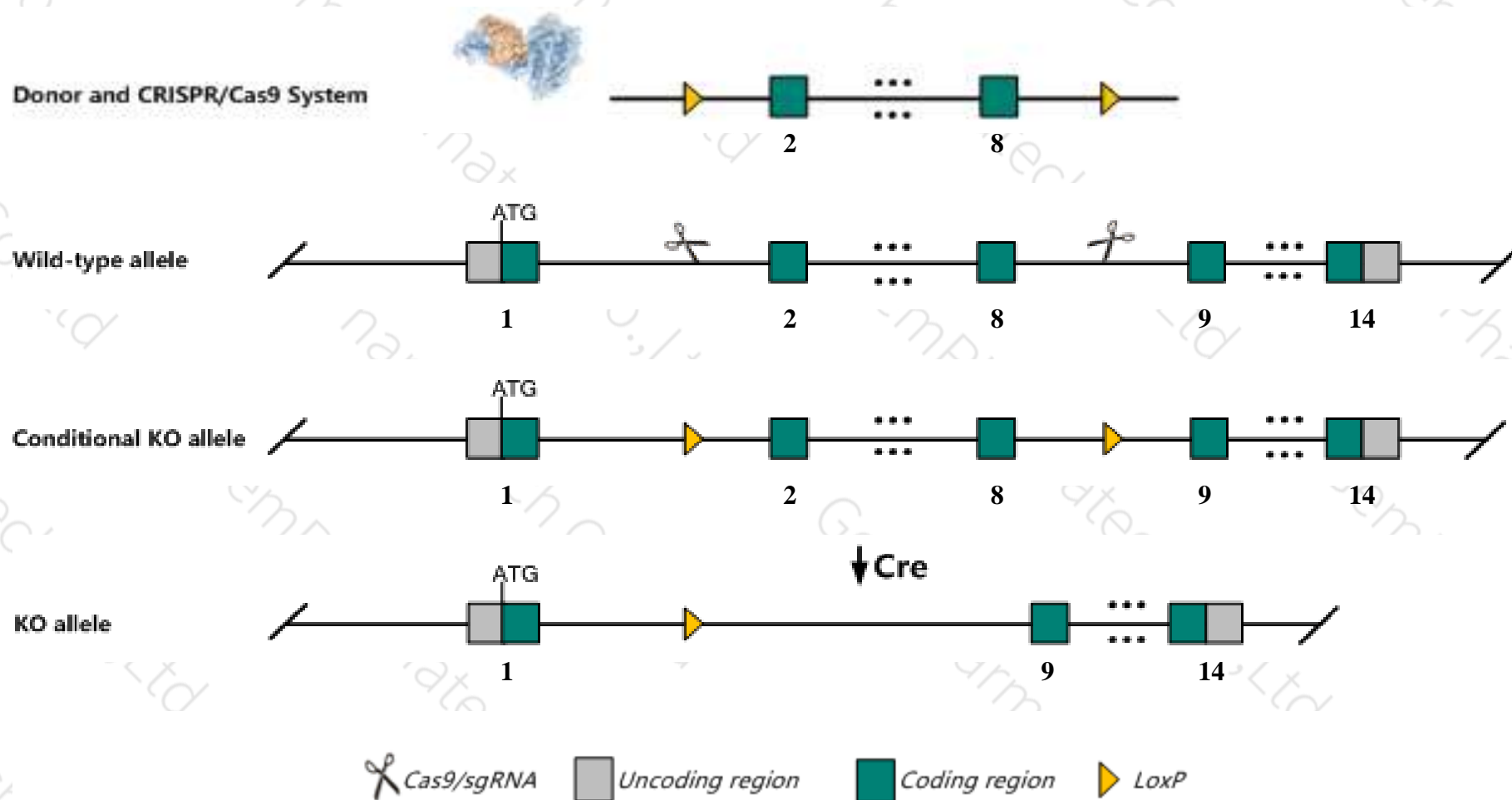
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kif22* gene. The schematic diagram is as follows:



Technical routes

- The *Kif22* gene has 6 transcripts. According to the structure of *Kif22* gene, exon2-exon8 of *Kif22*-201(ENSMUST00000032915.7) transcript is recommended as the knockout region. The region contains 1207bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Kif22* 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.

- According to the existing MGI data, mice homozygous for a null allele exhibit embryonic lethality prior to implantation due to defective meiosis II and early embryo mitosis.
- The *Kif22* 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.
- 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)

Kif22 kinesin family member 22 [Mus musculus (house mouse)]

Gene ID: 110033, updated on 13-Mar-2020

Summary



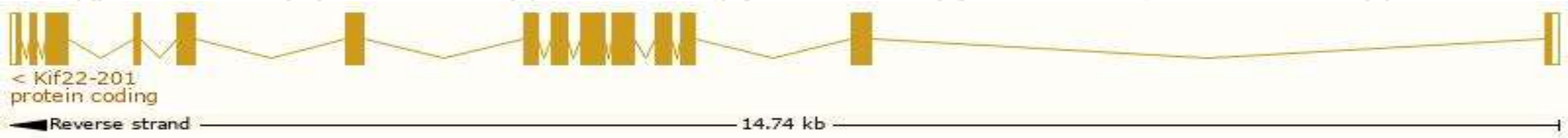
Official Symbol	Kif22 provided by MGI
Official Full Name	kinesin family member 22 provided by MGI
Primary source	MGI:MGI:109233
See related	Ensembl:ENSMUSG00000030677
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AU021460, C81217, Kid, Kif22a
Expression	Biased expression in CNS E11.5 (RPKM 35.8), liver E14 (RPKM 33.5) and 13 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

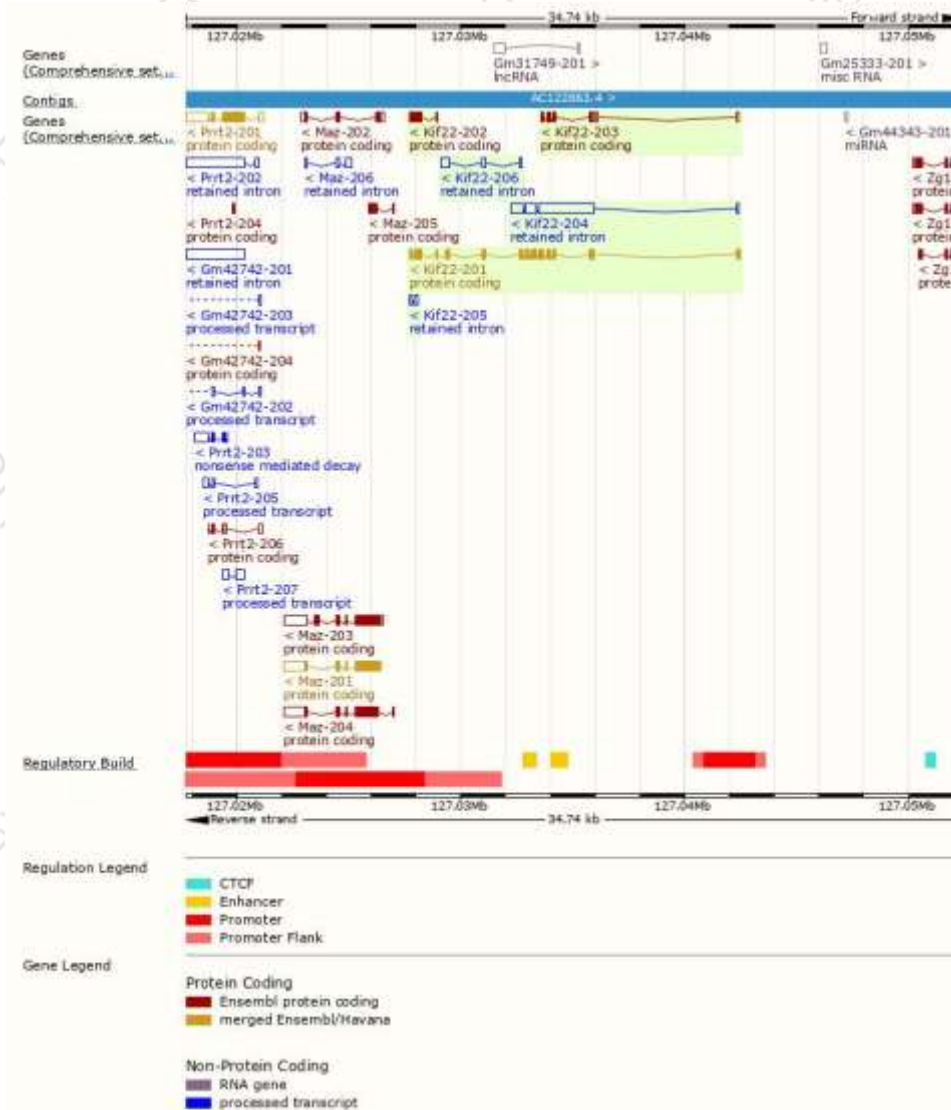
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kif22-201	ENSMUST00000032915.7	2115	660aa	Protein coding	CCDS21855	Q3V300	TSL:1 GENCODE basic APPRIS P1
Kif22-203	ENSMUST00000205806.1	706	161aa	Protein coding	-	A0A0U1RPH8	CDS 3' incomplete TSL:3
Kif22-202	ENSMUST00000205754.1	518	151aa	Protein coding	-	A0A0U1RNY7	CDS 5' incomplete TSL:3
Kif22-204	ENSMUST00000206412.1	3586	No protein	Retained intron	-	-	TSL:1
Kif22-206	ENSMUST00000206924.1	642	No protein	Retained intron	-	-	TSL:2
Kif22-205	ENSMUST00000206655.1	295	No protein	Retained intron	-	-	TSL:2

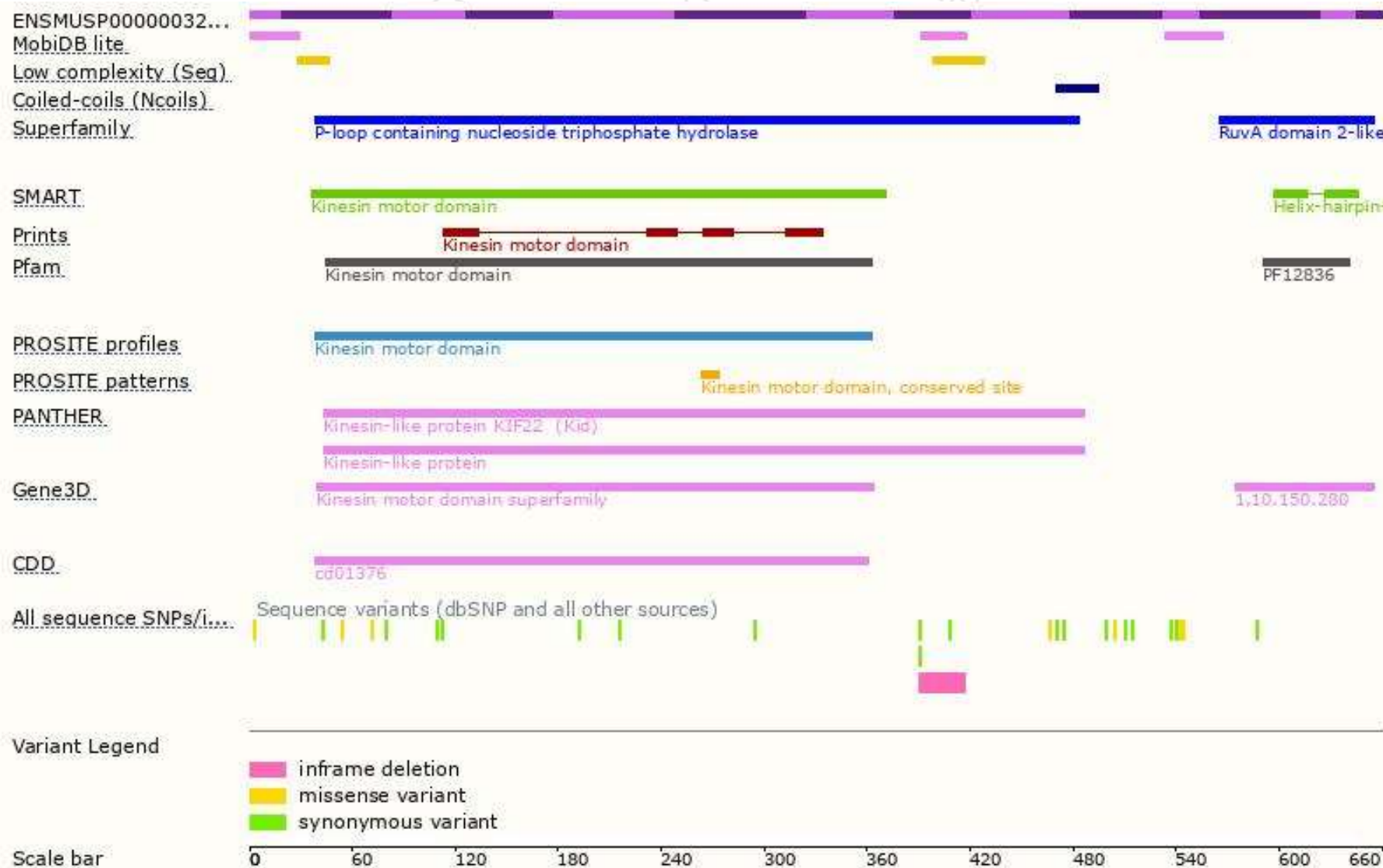
The strategy is based on the design of *Kif22-201* transcript,the transcription is shown below:



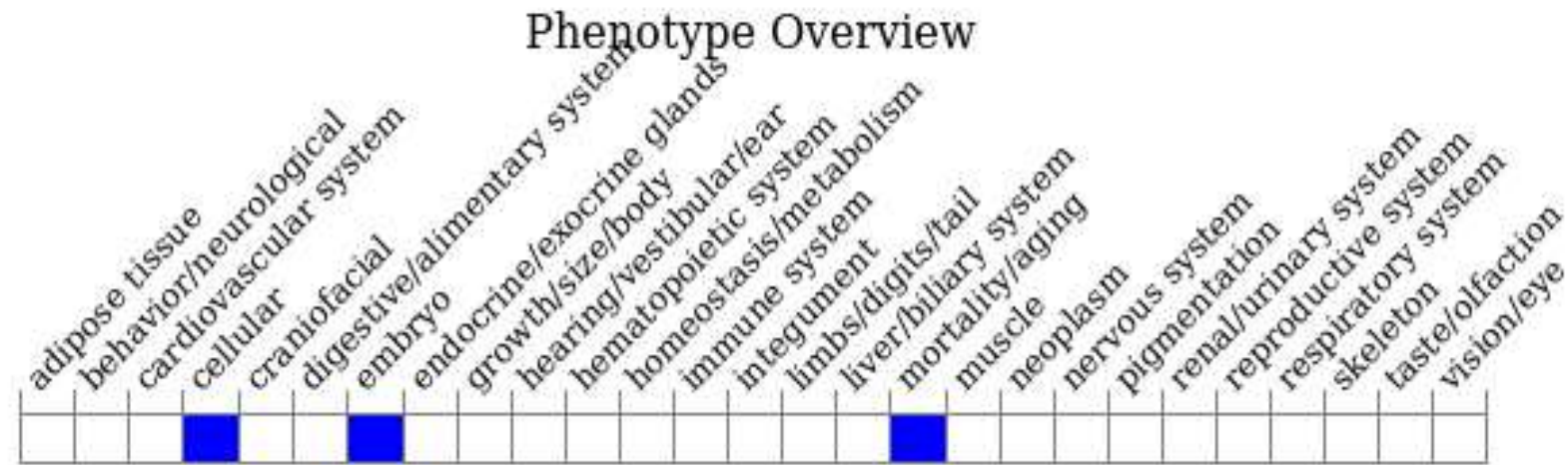
Genomic location distribution



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, mice homozygous for a null allele exhibit embryonic lethality prior to implantation due to defective meiosis II and early embryo mitosis.

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

Tel: 025-5864 1534

