Kpnb1 Cas9-CKO Strategy

Designer: Bingxuan Li

Design Date: 2019-9-5

Project Overview



Project Name

Kpnb1

Project type

Cas9-CKO

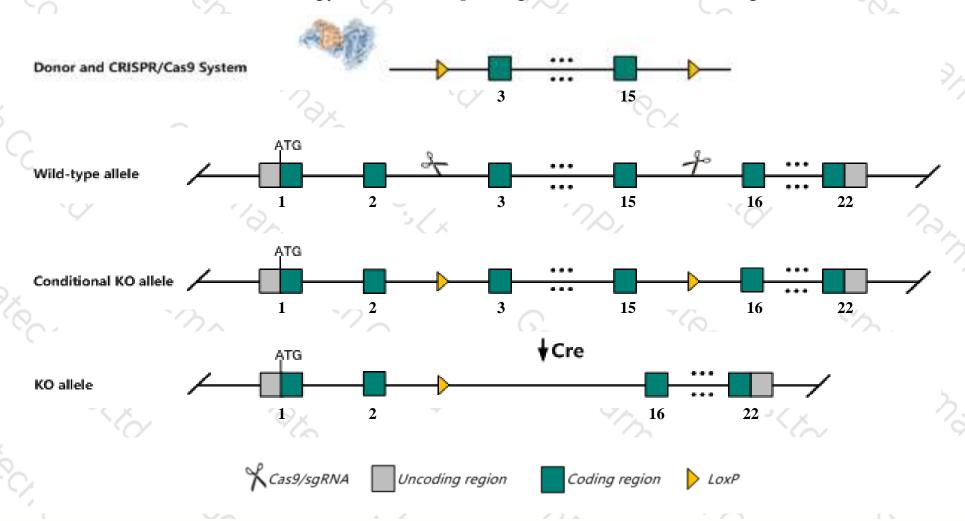
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Kpnb1* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Kpnb1* gene has 3 transcripts. According to the structure of *Kpnb1* gene, exon3-exon15 of *Kpnb1-201* (ENSMUST0000001479.4) transcript is recommended as the knockout region. The region contains 1813bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Kpnb1* 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



- > According to the existing MGI data, Homozygous mutation of this gene results in lethality shortly after implantation.
- ➤ The *Kpnb1* gene is located on the Chr11. 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)



Kpnb1 karyopherin (importin) beta 1 [Mus musculus (house mouse)]

Gene ID: 16211, updated on 12-Aug-2019

Summary



Official Symbol Kpnb1 provided by MGI

Official Full Name karyopherin (importin) beta 1 provided by MGI

Primary source MGI:MGI:107532

See related Ensembl: ENSMUSG00000001440

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 IPOB; Impnb; AA409963

Expression Ubiquitous expression in CNS E11.5 (RPKM 73.8), placenta adult (RPKM 41.1) and 26 other tissues See more

Orthologs human all

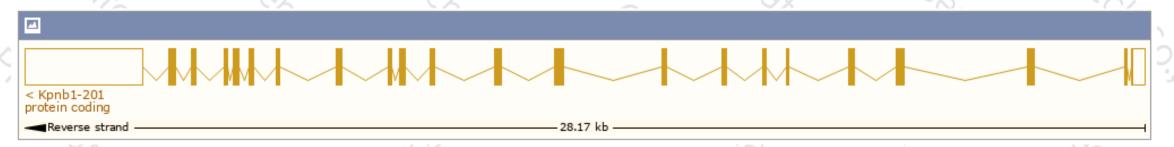
Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

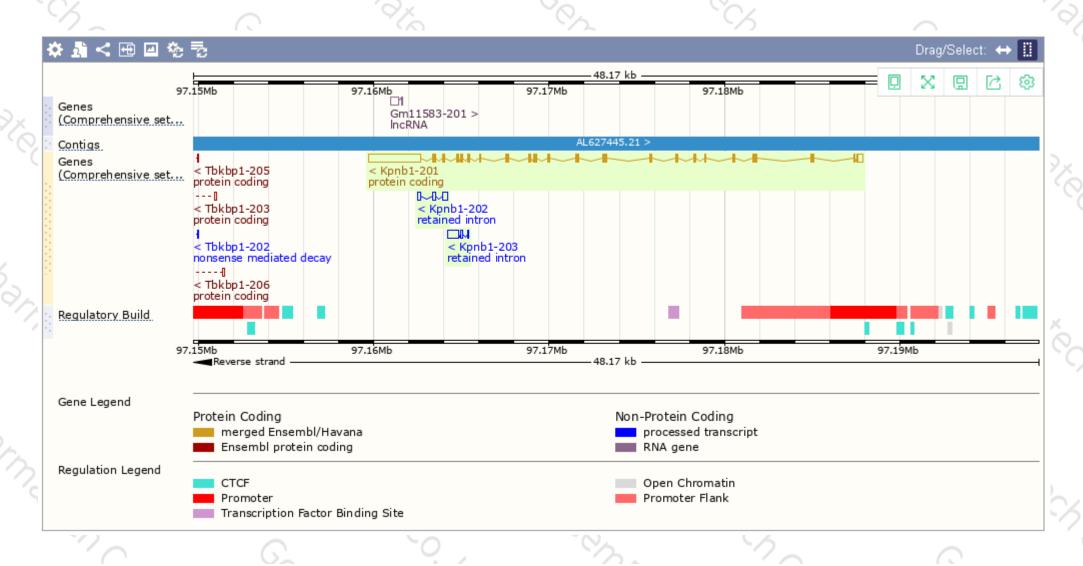
Show/hide columns (1 hidden)								Filter	X C
Name 🌲	Transcript ID 🔺	bp 🌲	Protein 🌲	Biotype 🌲	CCDS	UniProt 🌲		Flags	
Kpnb1-201	ENSMUST00000001479.4	5894	<u>876aa</u>	Protein coding	<u>CCDS25316</u> ₽	<u>P70168</u> @	TSL:1	GENCODE basic	APPRIS P1
Kpnb1-202	ENSMUST00000132633.1	643	No protein	Retained intron	-	-		TSL:2	
Kpnb1-203	ENSMUST00000153727.2	779	No protein	Retained intron	-	-		TSL:2	

The strategy is based on the design of *Kpnb1-201* transcript, The transcription is shown below:



Genomic location distribution





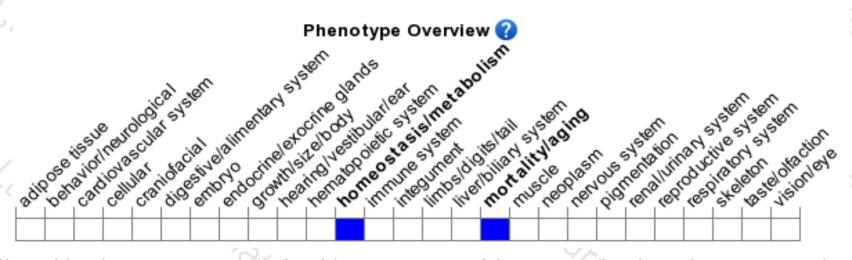
Protein domain





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, Homozygous mutation of this gene results in lethality shortly after implantation.



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

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