

Prkdc Cas9-CKO Strategy

Designer: Yupeng Yang

Reviewer: Shilei Zhu

Date: 2018/11/12

Project Overview



Project Name

Prkdc

Project type

Cas9-CKO

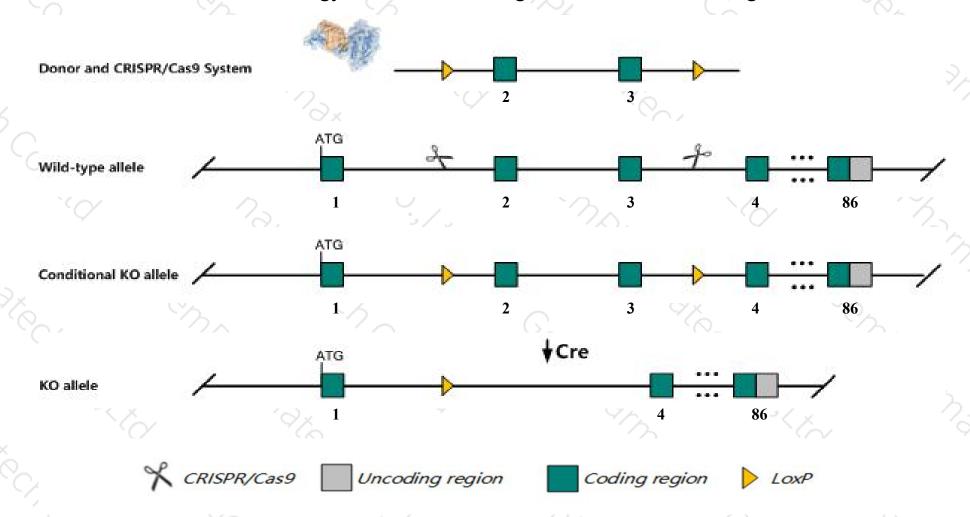
Strain background

C57BL/6JGpt

Conditional Knockout strategy



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



Technical routes



- The *Prkdc* gene has 2 transcripts. According to the structure of *Prkdc* gene, exon2-exon3 of *Prkdc-201* (ENSMUST00000023352.8) transcript is recommended as the knockout region. The region contains 170bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Prkdc* 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, Mutations at this locus effect genome stability, radiation sensitivity and DNA repair. Nonsense (scid) and null homozygotes have severe combined immunodeficiency. A BALB/c variant allele reduces enzyme activity and predisposes to breast cancer.
- > The *Prkdc* gene is located on the Chr16. 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)



Prkdc protein kinase, DNA activated, catalytic polypeptide [Mus musculus (house mouse)]

Gene ID: 19090, updated on 9-Apr-2019

Summary

☆ ?

Official Symbol Prkdc provided by MGI

Official Full Name protein kinase, DNA activated, catalytic polypeptide provided by MGI

Primary source MGI:MGI:104779

See related Ensembl:ENSMUSG00000022672

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 Al326420, AU019811, DNA-PKcs, DNAPDcs, DNAPK, DNPK1, DOXNPH, HYRC1, XRCC7, dxnph, p460, scid, slip

Expression Ubiquitous expression in CNS E11.5 (RPKM 1.5), frontal lobe adult (RPKM 1.2) and 27 other tissuesSee more

Orthologs <u>human</u> all

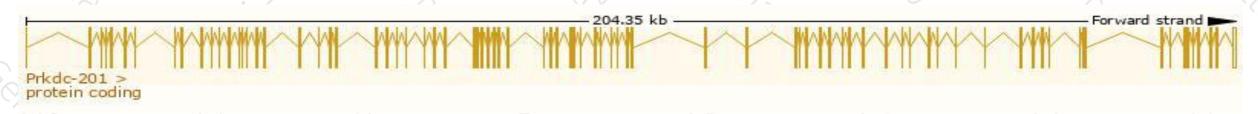
Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

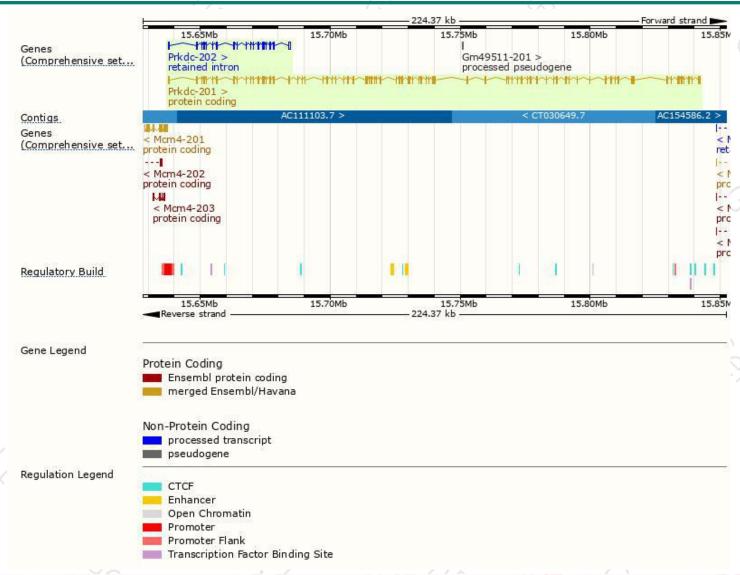
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Prkdc-201	ENSMUST00000023352.8	12647	4128aa	Protein coding	CCDS27978	P97313	TSL:1 GENCODE basic APPRIS P1
Prkdc-202	ENSMUST00000182134.1	3063	No protein	Retained intron	691		TSL:5

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



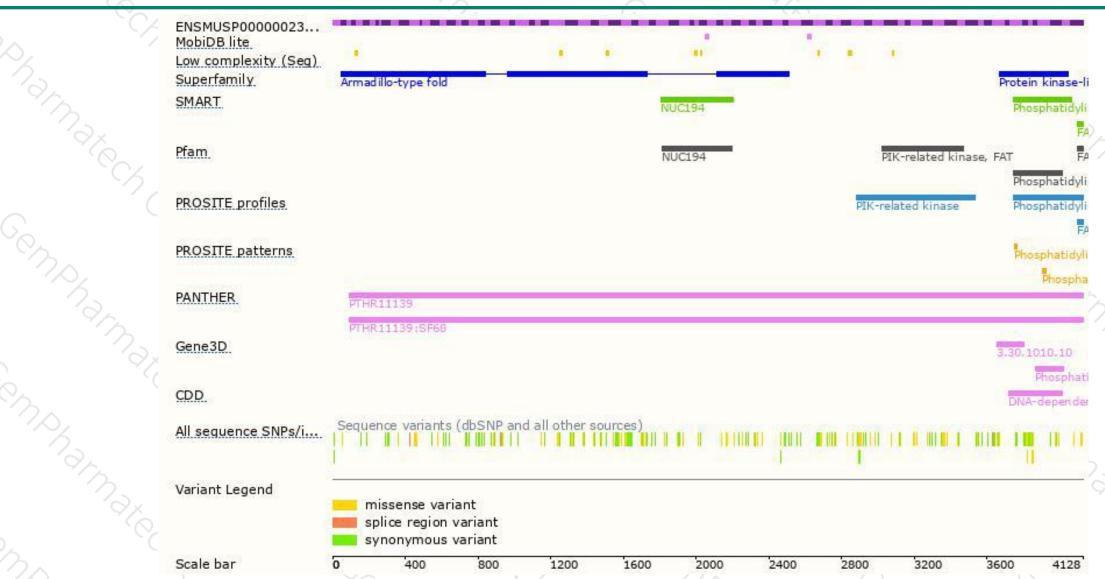
Genomic location distribution





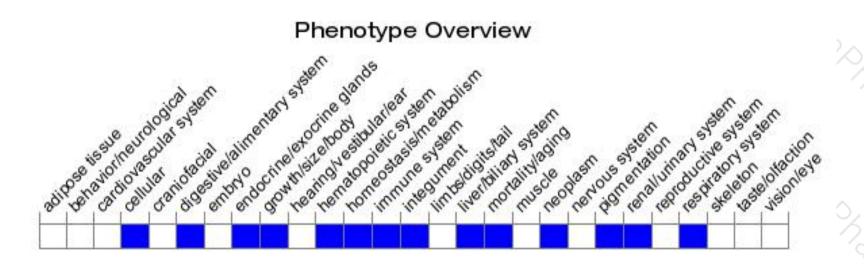
Protein domain





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, Mutations at this locus effect genome stability, radiation sensitivity and DNA repair. Nonsense (scid) and null homozygotes have severe combined immunodeficiency. A BALB/c variant allele reduces enzactivity and predisposes to breast cancer.



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





