Ptx3 Cas9-CKO Strategy

Designer: Daohua Xu

Design Date: 2019-10-22

Project Overview



Project Name

Ptx3

Project type

Cas9-CKO

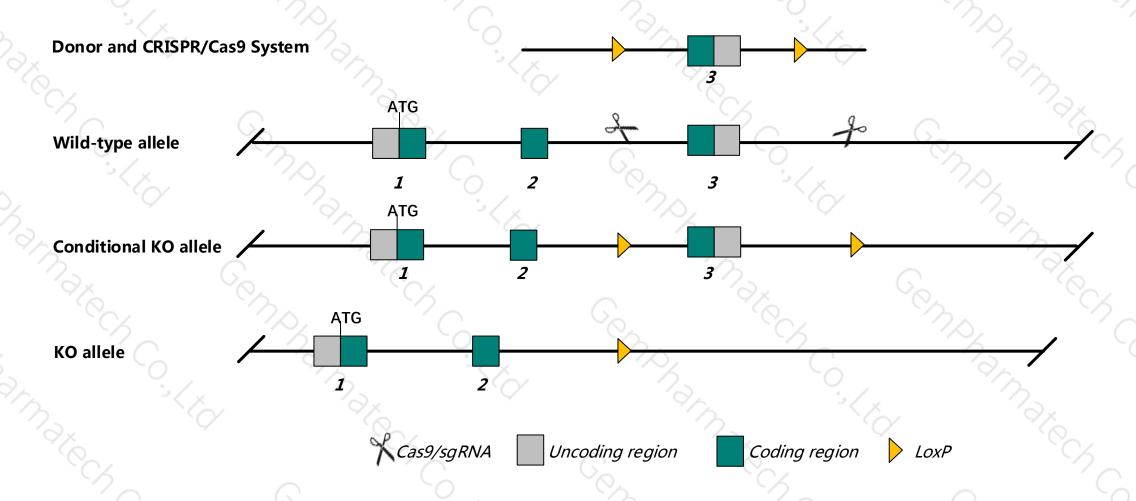
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ptx3* gene. The schematic diagram is as follows:



Technical routes



- The *Ptx3* gene has 2 transcripts. According to the structure of *Ptx3* gene, exon3 of *Ptx3*-201 (ENSMUST00000029421.5) transcript is recommended as the knockout region. The region contains key coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ptx3* 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 or cell types.

Notice



- According to the existing MGI data, Homozygous mutant mice display female subfertility due to abnormalities of the cumulus oophorus and are susceptible to invasive pulmonary aspergillosis associated with defective recognition of conidia by alveolar macrophages and dendritic cells and impaired induction of adaptive type 2 responses.
- > The KO region contains functional region of the *Veph1* gene.Knockout the region may affect the function of *Veph1* gene.
- The *Ptx3* gene is located on the Chr3. 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Ptx3 pentraxin related gene [Mus musculus (house mouse)]

Gene ID: 19288, updated on 12-Aug-2018

Summary

☆ ?

Official Symbol Ptx3 provided by MGI

Official Full Name pentraxin related gene provided by MGI

Primary source MGI:MGI:104641

See related Ensembl:ENSMUSG00000027832 Vega:OTTMUSG00000043511

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 TSG-14; Al607804

Expression Biased expression in limb E14.5 (RPKM 16.3), CNS E11.5 (RPKM 14.4) and 7 other tissues See more

Orthologs <u>human</u> all

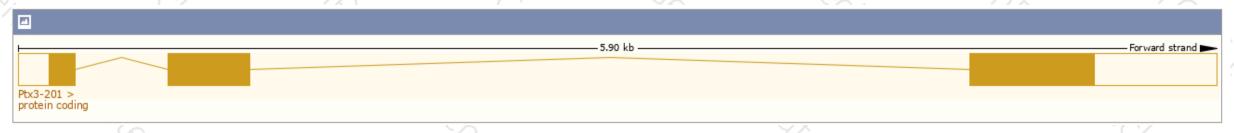
Transcript information (Ensembl)



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

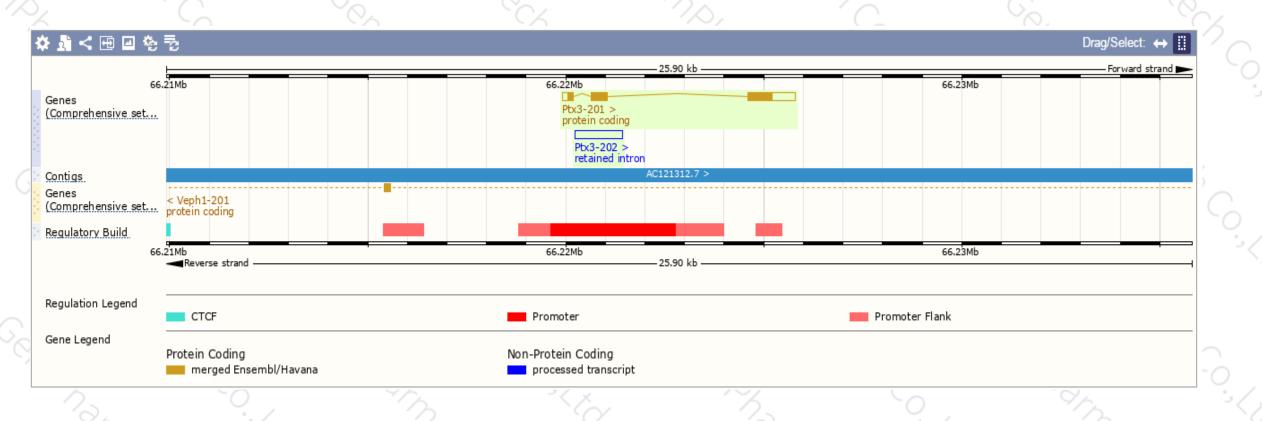
| Show/hide columns (1 hidden) | | | | | | | | Filter | |
|------------------------------|----------------------|------|--------------|-----------------|--------------------|-----------------|------------------------|---------------------|-----------|
| Name 🍦 | Transcript ID | bp 🌲 | Protein | Biotype | CCDS 🍦 | UniProt 🍦 | RefSeq | Flags | * |
| Ptx3-201 | ENSMUST00000029421.5 | 1898 | <u>381aa</u> | Protein coding | <u>CCDS17392</u> & | <u>P48759</u> ₽ | NM_008987 NP_033013 | TSL:1 GENCODE basic | APPRIS P1 |
| Ptx3-202 | ENSMUST00000182521.1 | 1186 | No protein | Retained intron | - | - | - | TSL:NA | |

The strategy is based on the design of *Ptx3*-201 transcript, The transcription is shown below



Genomic location distribution





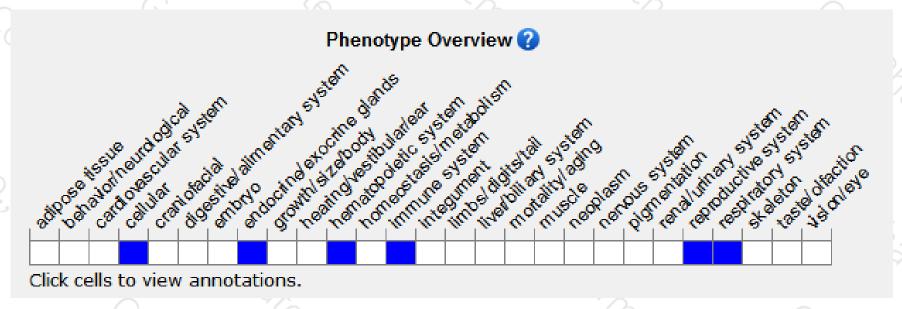
Protein domain





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 mutant mice display female subfertility due to abnormalities of the cumulus oophorus and are susceptible to invasive pulmonary aspergillosis associated with defective recognition of conidia by alveolar macrophages and dendritic cells and impaired induction of adaptive type 2 responses.

If you have any questions, you are welcome to inquire. Tel: 025-5864 1534





