

Pex10 Cas9-CKO Strategy

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Reviewer: Lingyan Wu

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Project Overview



Project Name Pex10

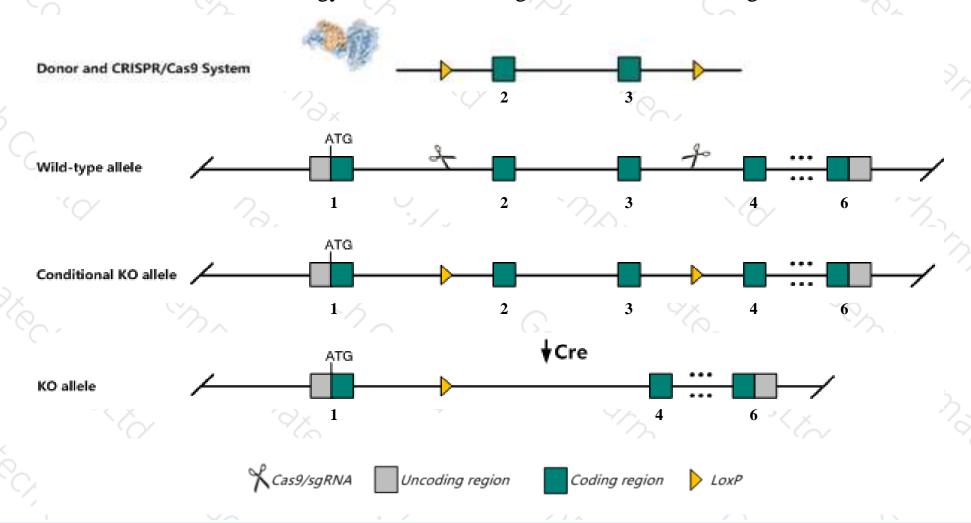
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Pex10* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Pex10* gene has 5 transcripts. According to the structure of *Pex10* gene, exon2-exon3 of *Pex10*-201(ENSMUST00000103180.3) transcript is recommended as the knockout region. The region contains 488bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Pex10* 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, mice homozygous for an ENU-induced allele exhibit partial neonatal mortality due to respiratory distress, loss of embryonic movement, and prenatal pathology including altered biochemistry, defects in axonal integrity, decreased Schwann cell number, and defects at the neuromuscular junction.
- \succ The *Pex10* 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)



Pex10 peroxisomal biogenesis factor 10 [Mus musculus (house mouse)]

Gene ID: 668173, updated on 13-Mar-2020

Summary

A ?

Official Symbol Pex10 provided by MGI

Official Full Name peroxisomal biogenesis factor 10 provided by MGI

Primary source MGI:MGI:2684988

See related Ensembl:ENSMUSG00000029047

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 AV128229, Gm142

Expression Ubiquitous expression in adrenal adult (RPKM 12.3), ovary adult (RPKM 11.7) and 28 other tissuesSee more

Orthologs <u>human all</u>

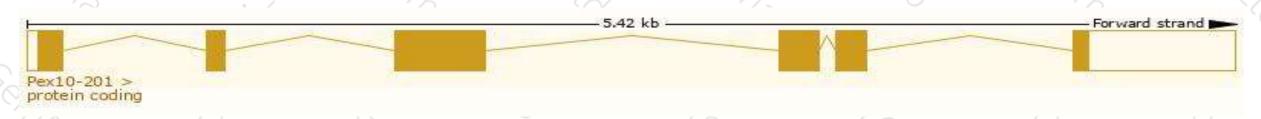
Transcript information (Ensembl)



The gene has 5 transcripts, all transcripts are shown below:

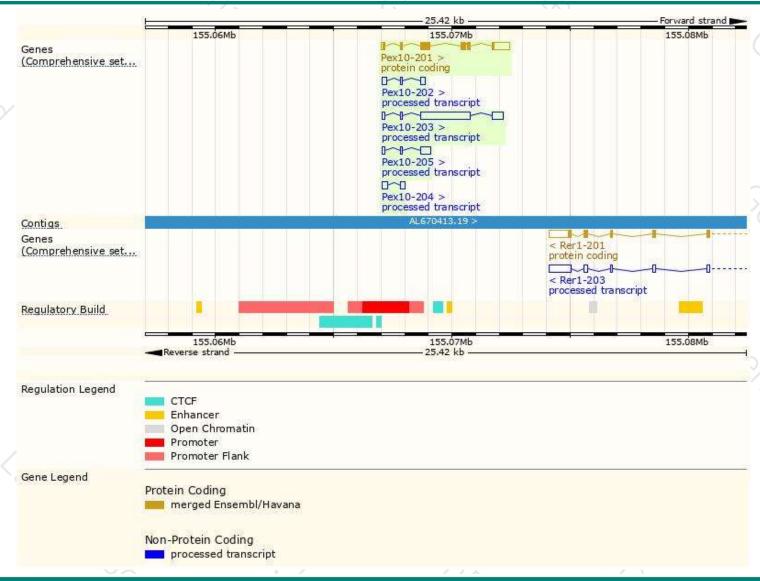
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pex10-201	ENSMUST00000103180.3	1688	324aa	Protein coding	CCDS19021	B1AUE5	TSL:1 GENCODE basic APPRIS P1
Pex10-203	ENSMUST00000125432.7	2790	No protein	Processed transcript	28	14	TSL:1
Pex10-205	ENSMUST00000134341.1	651	No protein	Processed transcript	100	24	TSL:2
Pex10-202	ENSMUST00000123395.7	491	No protein	Processed transcript			TSL:3
Pex10-204	ENSMUST00000133116.1	412	No protein	Processed transcript	¥	64	TSL:3

The strategy is based on the design of Pex10-201 transcript, the transcription is shown below:



Genomic location distribution





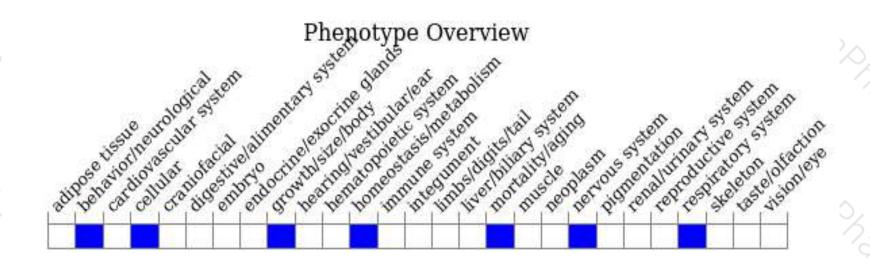
Protein domain





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 an ENU-induced allele exhibit partial neonatal mortality due to respiratory distress, loss of embryonic movement, and prenatal pathology including altered biochemistry, defects in axonal integrity, decreased Schwann cell number, and defects at the neuromuscular junction.



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

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