

Cyp11a1 Cas9-CKO Strategy

Designer:

Huan Fan

Design Date:

2019-7-25

Project Overview



Project Name

Cyp11a1

Project type

Cas9-CKO

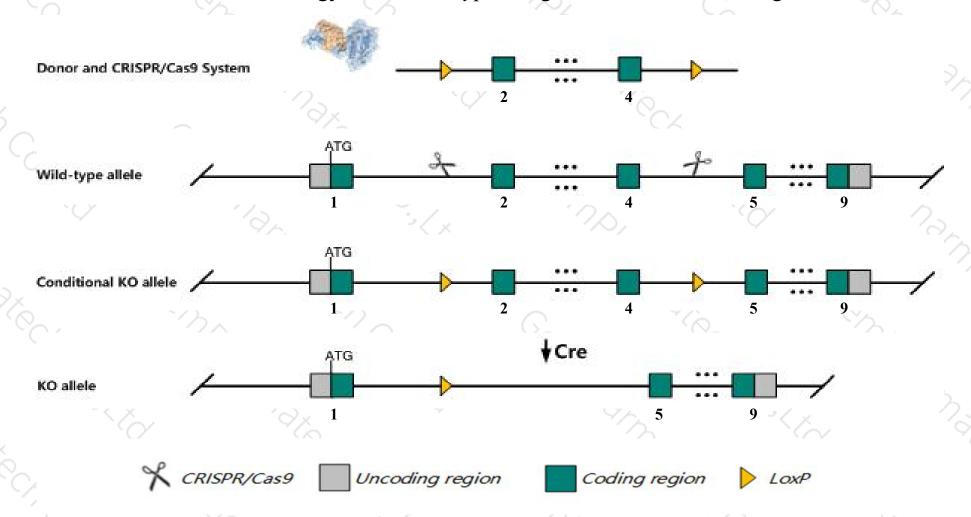
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cyp11a1* gene. The schematic diagram is as follows:



Technical routes



- The *Cyp11a1* gene has 4 transcripts. According to the structure of *Cyp11a1* gene, exon2-exon4 of *Cyp11a1-201* (ENSMUST00000034874.13) transcript is recommended as the knockout region. The region contains 560bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cyp11a1* 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, Homozygous null mice are exhibit a steroid deficiency and die within days of birth showing signs of dehydration. Males are feminized with female external genitalia and underdeveloped gonads. Mice homozgyous for another knock-out allele exhibit abnormal adrenal development and neonatal lethality.
- The *Cyp11a1* gene is located on the Chr9. 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)



Cyp11a1 cytochrome P450, family 11, subfamily a, polypeptide 1 [Mus musculus (house mouse)]

Gene ID: 13070, updated on 19-Mar-2019

Summary

☆ ?

Official Symbol Cyp11a1 provided by MGI

Official Full Name cytochrome P450, family 11, subfamily a, polypeptide 1 provided by MGI

Primary source MGI:MGI:88582

See related Ensembl: ENSMUSG00000032323

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 Cyp11a, Cypxia1, D9Ertd411e, P450scc, Scc, cscc

Expression Biased expression in adrenal adult (RPKM 494.9), ovary adult (RPKM 389.8) and 1 other tissueSee more

Orthologs human all

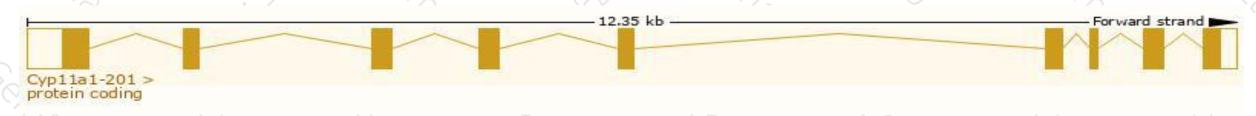
Transcript information (Ensembl)



The gene has 4 transcripts, all transcripts are shown below:

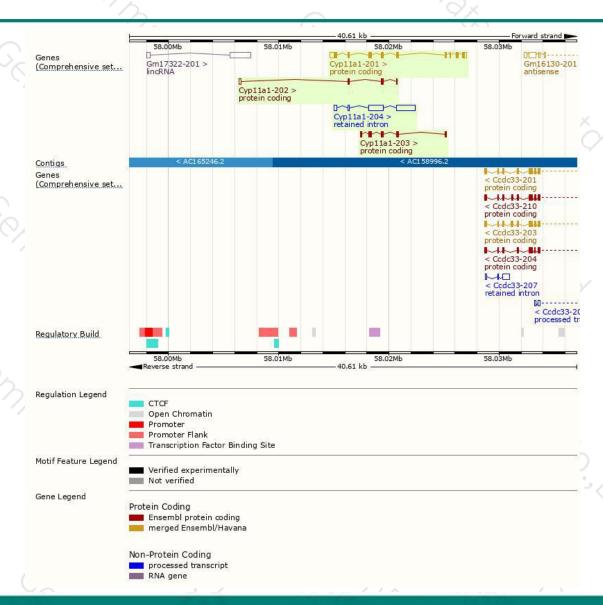
| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|-------------|-----------------------|------|--------------|-----------------|-----------|------------|-------------------------------|
| Cyp11a1-201 | ENSMUST00000034874.13 | 2120 | 526aa | Protein coding | CCDS40653 | Q9QZ82 | TSL:1 GENCODE basic APPRIS P1 |
| Cyp11a1-203 | ENSMUST00000188539.1 | 848 | 279aa | Protein coding | | A0A087WRU6 | CDS 3' incomplete TSL:2 |
| Cyp11a1-202 | ENSMUST00000188116.6 | 539 | <u>115aa</u> | Protein coding | 14 | A0A087WRA8 | CDS 3' incomplete TSL:3 |
| Cyp11a1-204 | ENSMUST00000188944.1 | 3439 | No protein | Retained intron | 1 4 | 828 | TSL:1 |

The strategy is based on the design of Cyp11a1-201 transcript, The transcription is shown below



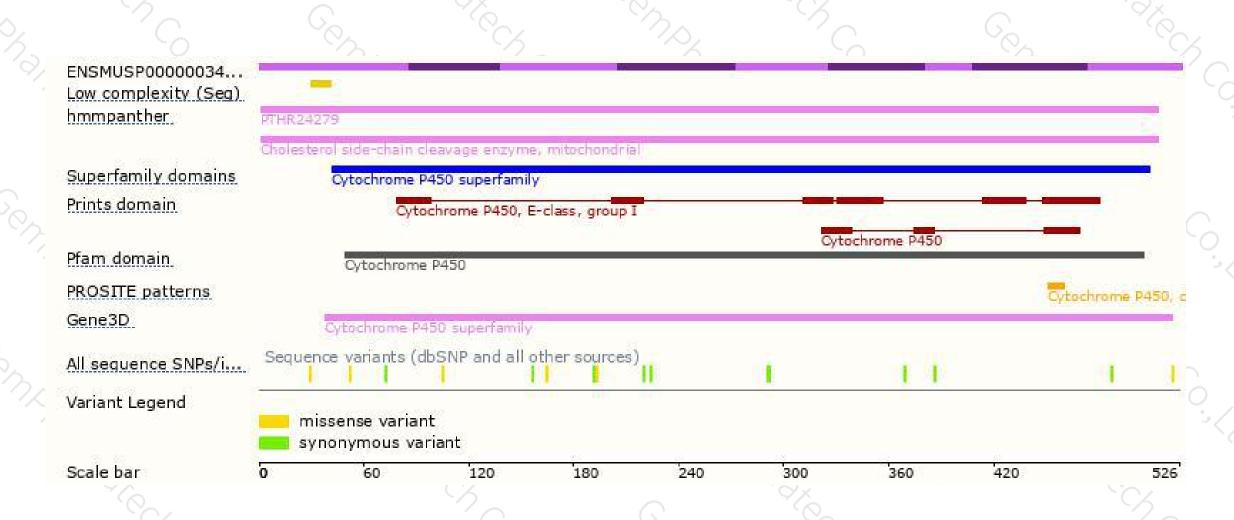
Genomic location distribution





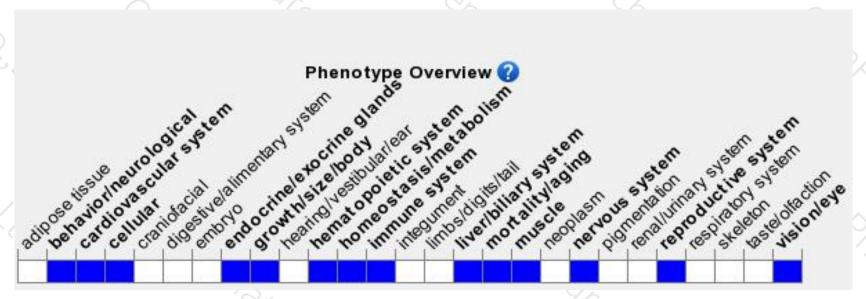
Protein domain





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 null mice are exhibit a steroid deficiency and die within days of birth showing signs of dehydration. Males are feminized with female external genitalia and underdeveloped gonads. Mice homozgyous for another knock-out allele exhibit abnormal adrenal development and neonatal lethality.



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





