Nr1i3 Cas9-CKO Strategy

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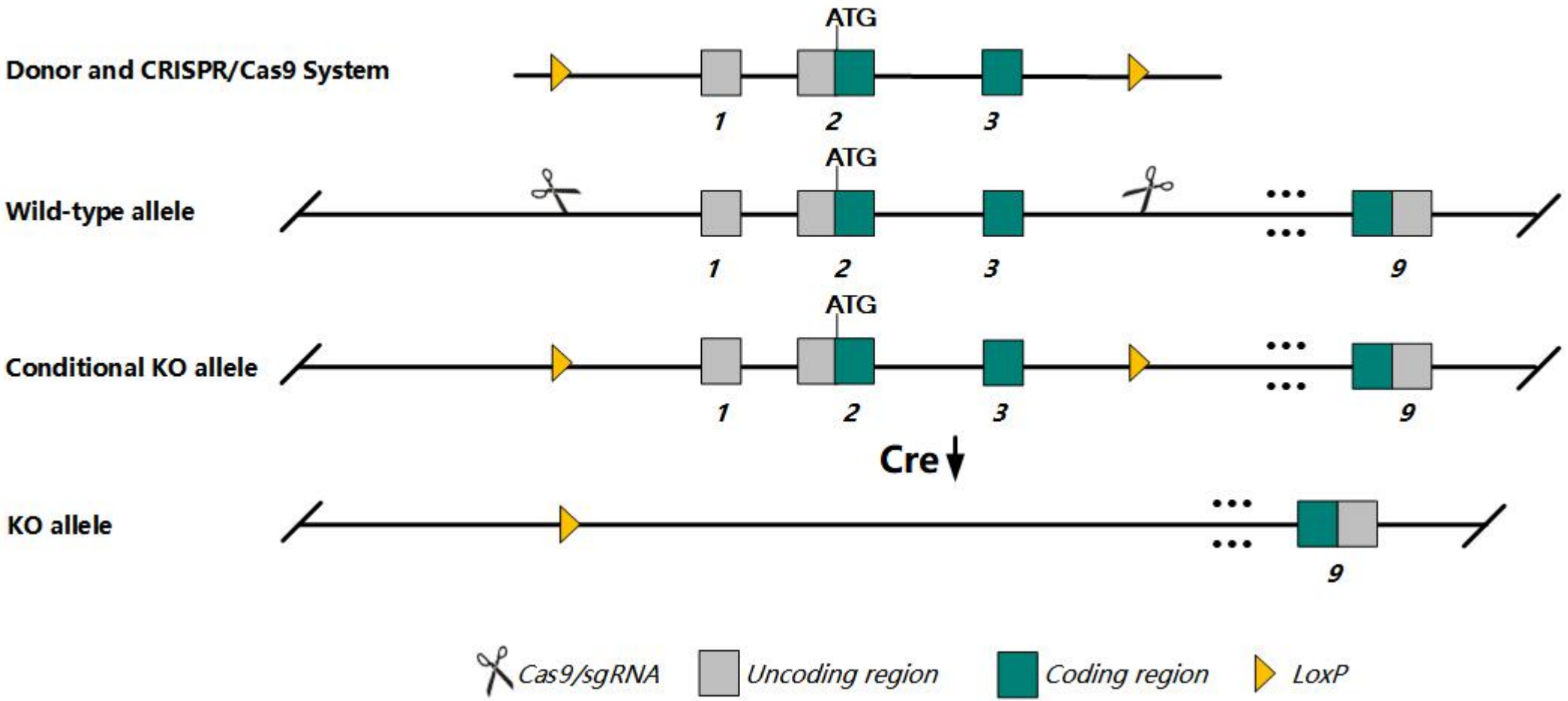
Project Overview

Project Name	***Nr1i3***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Nr1i3* gene. The schematic diagram is as follows:



Technical routes

- The *Nr1i3* gene has 8 transcripts. According to the structure of *Nr1i3* gene, exon1-exon3 of *Nr1i3-201* (ENSMUST00000005820.10) transcript is recommended as the knockout region.The region contains ATG coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Nr1i3* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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, mice homozygous for a knock-out allele exhibit decreased sensitivity to TCPOBOP.
- The *Tomm40l* gene may be affected.
- The *Nr1i3* gene is located on the Chr1. 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）

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Nr1i3 nuclear receptor subfamily 1, group I, member 3 [Mus musculus (house mouse)]

Gene ID: 12355, updated on 13-Mar-2020

Summary

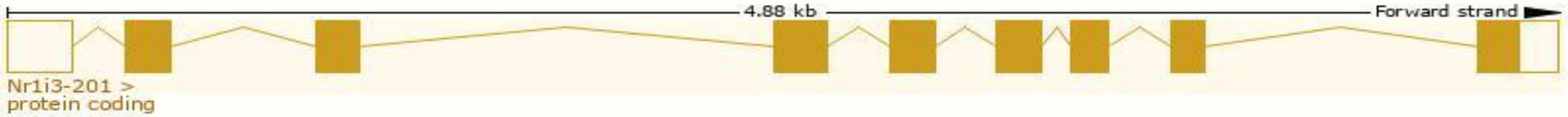
Official Symbol	Nr1i3 provided by MGI
Official Full Name	nuclear receptor subfamily 1, group I, member 3 provided by MGI
Primary source	MGI:MGI:1346307
See related	Ensembl:ENSMUSG00000005677
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AA209988, AI551208, CAR, CAR-beta, Care2, ESTM32, MB67
Expression	Biased expression in liver adult (RPKM 52.5), duodenum adult (RPKM 13.0) and 3 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

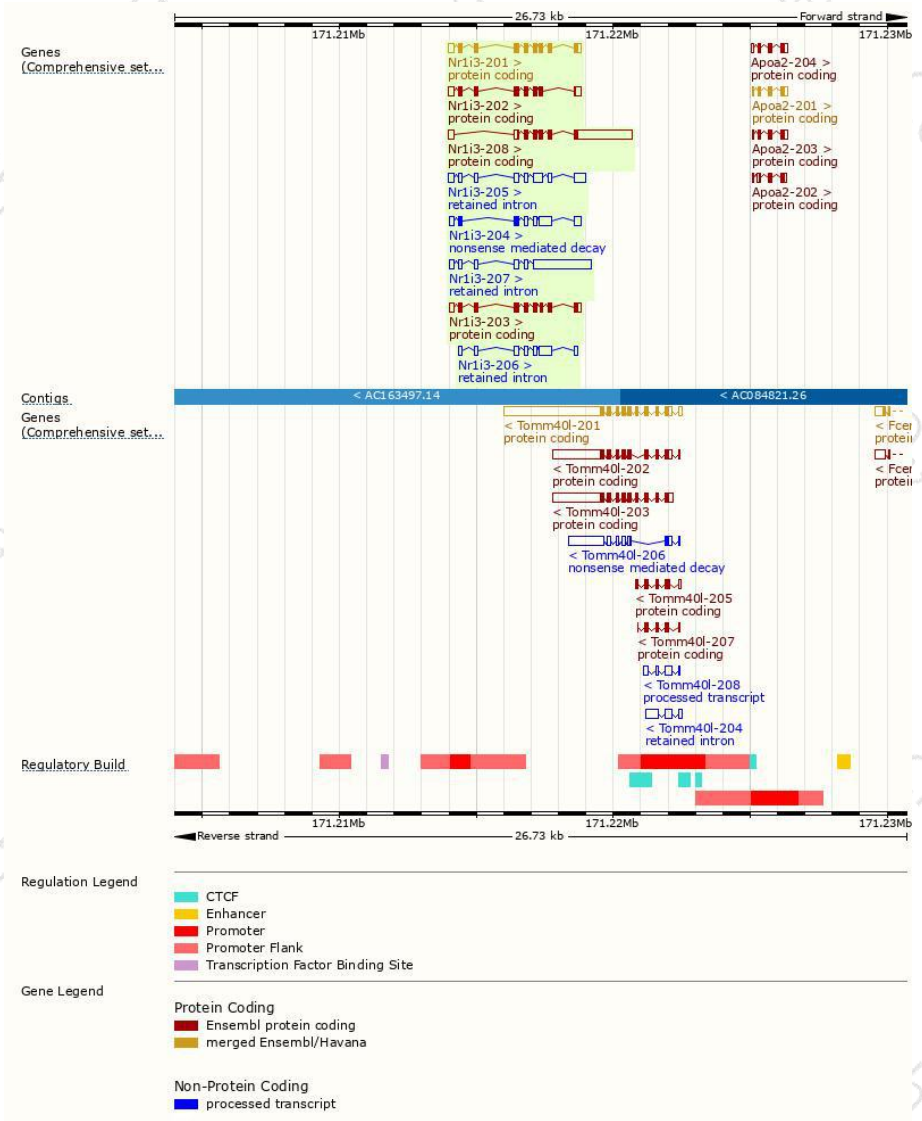
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nr1i3-201	ENSMUST00000005820.10	1414	358aa	Protein coding	CCDS15480	O35627	TSL:1 GENCODE basic APPRIS P3
Nr1i3-203	ENSMUST00000111328.7	1359	357aa	Protein coding	CCDS56655	Q5FW96	TSL:1 GENCODE basic APPRIS ALT2
Nr1i3-202	ENSMUST00000075469.11	1305	286aa	Protein coding	CCDS56654	O35627	TSL:1 GENCODE basic
Nr1i3-208	ENSMUST00000155126.7	2980	221aa	Protein coding	-	Q3UEP1	TSL:2 GENCODE basic
Nr1i3-204	ENSMUST00000133075.7	1446	95aa	Nonsense mediated decay	-	M0QWI8	TSL:1
Nr1i3-207	ENSMUST00000152865.7	2855	No protein	Retained intron	-	-	TSL:2
Nr1i3-205	ENSMUST00000137298.7	1638	No protein	Retained intron	-	-	TSL:5
Nr1i3-206	ENSMUST00000149404.1	1272	No protein	Retained intron	-	-	TSL:5

The strategy is based on the design of *Nr1i3-201* transcript,the transcription is shown below

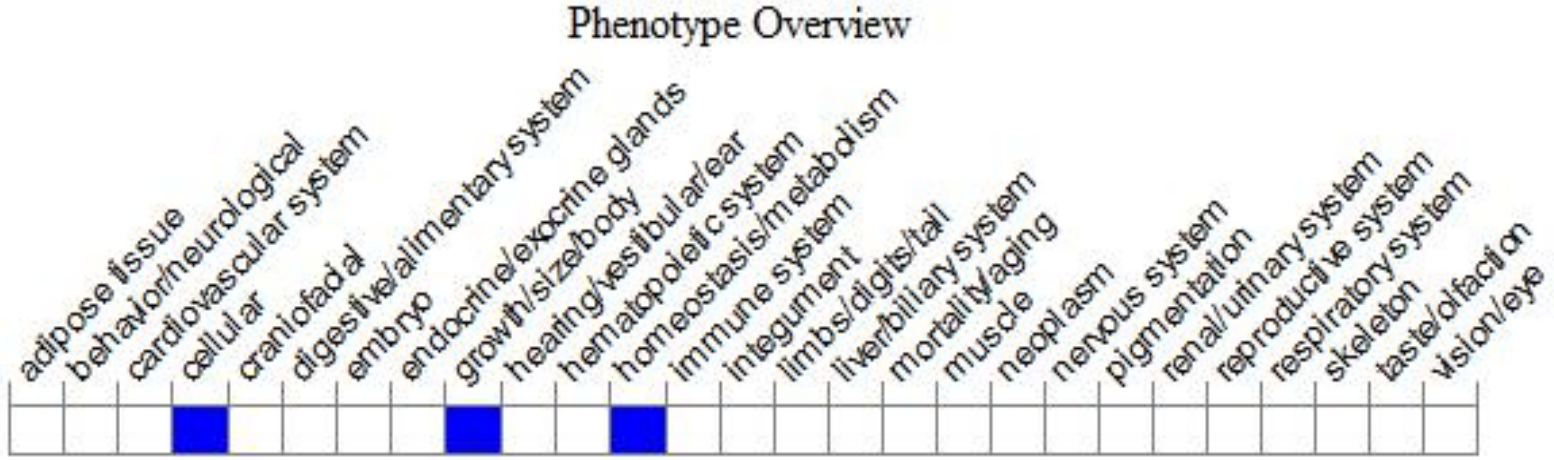


Genomic location distribution



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 a knock-out allele exhibit decreased sensitivity to TCPOBOP.

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

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