

Pofut1 Cas9-CKO Strategy

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Project Overview

Project Name

Pofut1

Project type

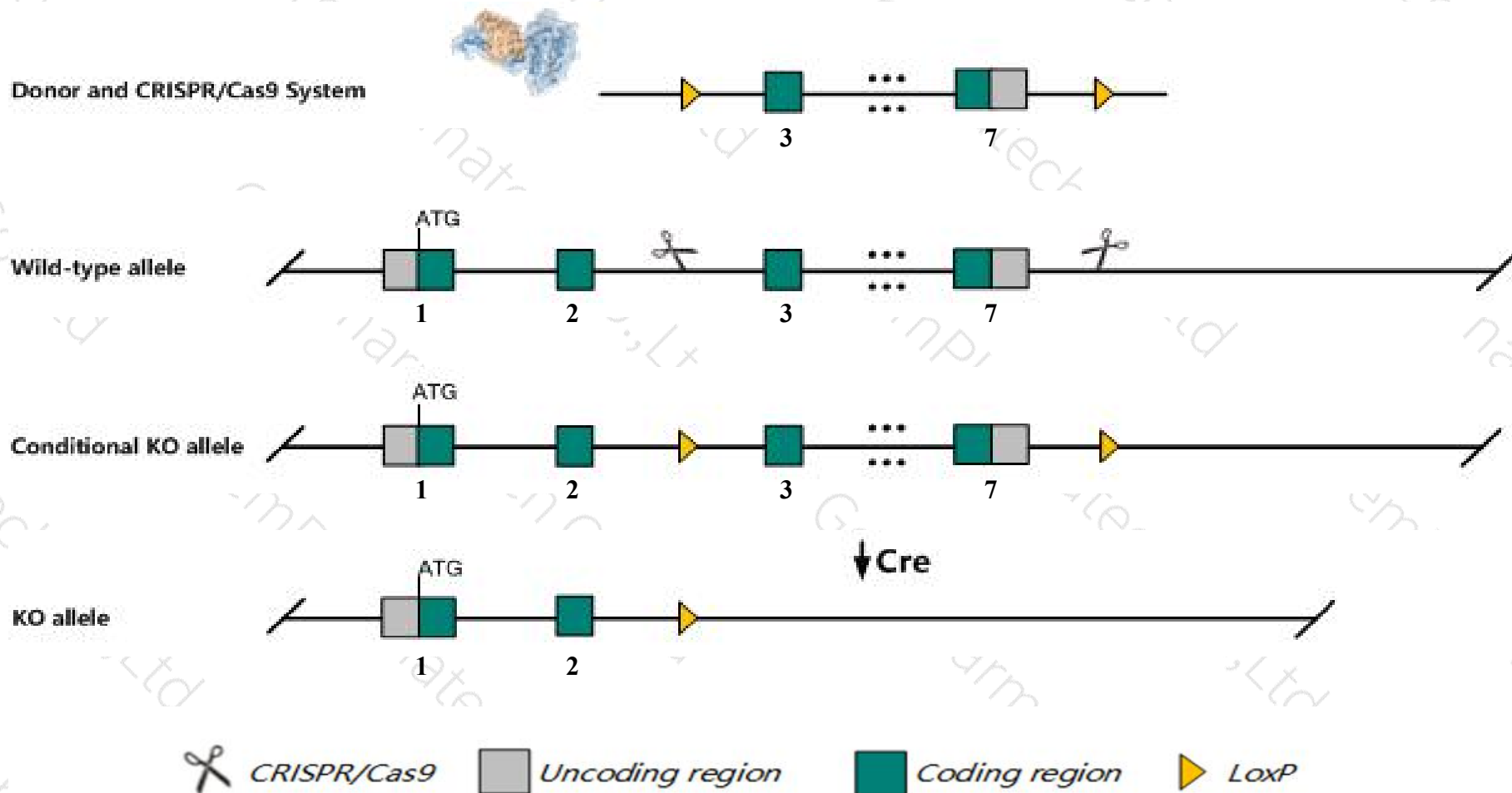
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pofut1* gene. The schematic diagram is as follows:



- The *Pofut1* gene has 8 transcripts. According to the structure of *Pofut1* gene, exon3-exon7 of *Pofut1-201* (ENSMUST00000049863.11) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Pofut1* 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.

- According to the existing MGI data, Homozygous mutant mice die by midgestation displaying malformations of the somites, vasculature, heart, and nervous system.
- The *Pofut1* gene is located on the Chr2. 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)

Pofut1 protein O-fucosyltransferase 1 [*Mus musculus* (house mouse)]

Gene ID: 140484, updated on 9-Feb-2020

Summary

Official Symbol	Pofut1 provided by MGI
Official Full Name	protein O-fucosyltransferase 1 provided by MGI
Primary source	MGI:MGI:2153207
See related	Ensembl:ENSMUSG00000046020
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	O-FucT-1; mKIAA0180
Expression	Ubiquitous expression in adrenal adult (RPKM 10.2), limb E14.5 (RPKM 10.0) and 28 other tissues See more
Orthologs	human all

Genomic context

Location: 2; 2 H1

Exon count: 8

See Pofut1 in [Genome Data Viewer](#)

Transcript information (Ensembl)

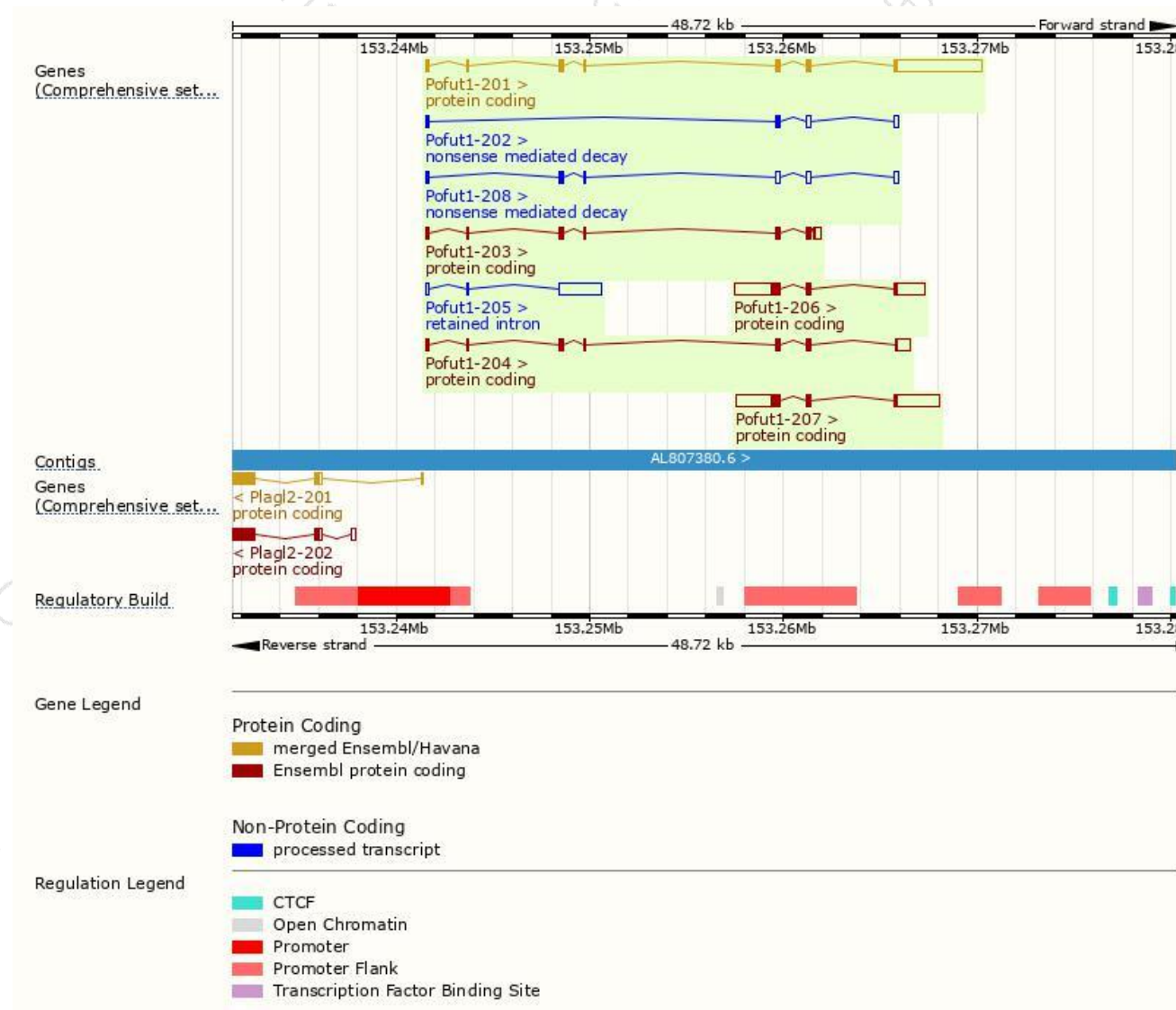
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pofut1-201	ENSMUST00000049863.11	5600	393aa	Protein coding	CCDS16909	Q91ZW2	TSL:1 GENCODE basic APPRIS P1
Pofut1-207	ENSMUST00000152390.1	4833	298aa	Protein coding	-	E9Q686	TSL:2 GENCODE basic
Pofut1-206	ENSMUST00000123487.1	4143	297aa	Protein coding	-	E9PZ15	TSL:2 GENCODE basic
Pofut1-204	ENSMUST00000109794.1	1688	352aa	Protein coding	-	A2AMC3	TSL:5 GENCODE basic
Pofut1-203	ENSMUST00000099192.9	1385	353aa	Protein coding	-	Q3UXG7	TSL:1 GENCODE basic
Pofut1-208	ENSMUST00000170297.7	1135	83aa	Nonsense mediated decay	-	E9Q154	TSL:5
Pofut1-202	ENSMUST00000099191.4	839	84aa	Nonsense mediated decay	-	E9Q7A1	TSL:5
Pofut1-205	ENSMUST00000123158.1	2413	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Pofut1-201* transcript,The transcription is shown below



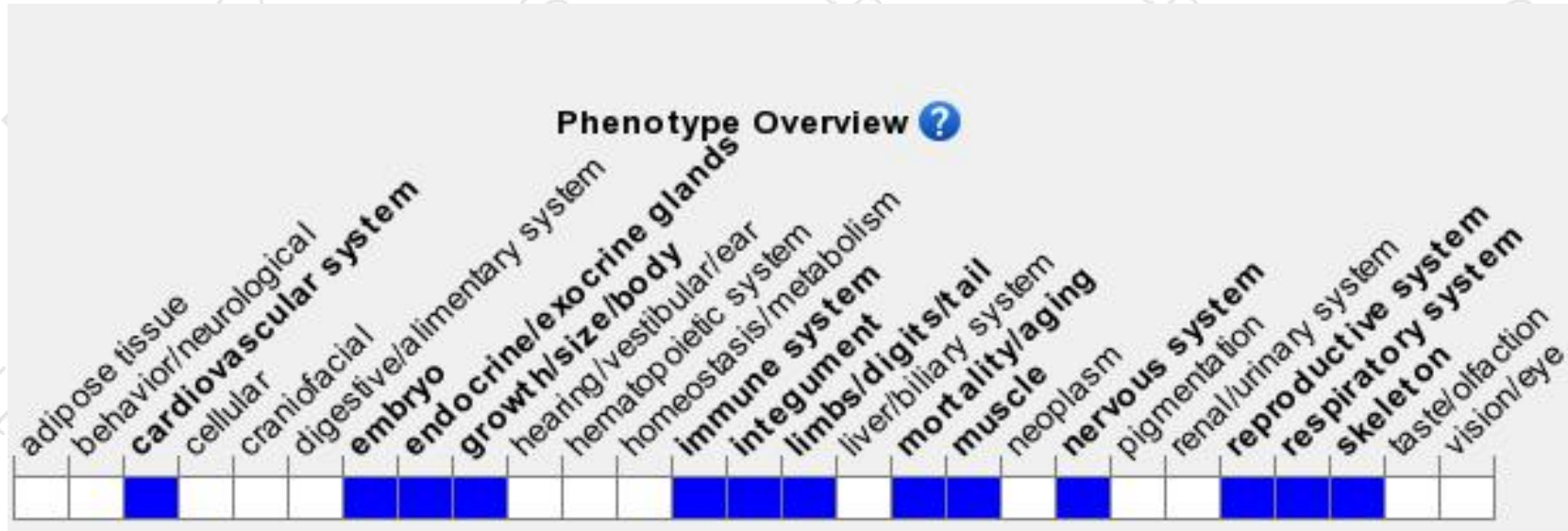
Genomic location distribution



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 die by midgestation displaying malformations of the somites, vasculature, heart, and nervous system.

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

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