

Pfas Cas9-CKO Strategy

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Project Overview

Project Name

Pfas

Project type

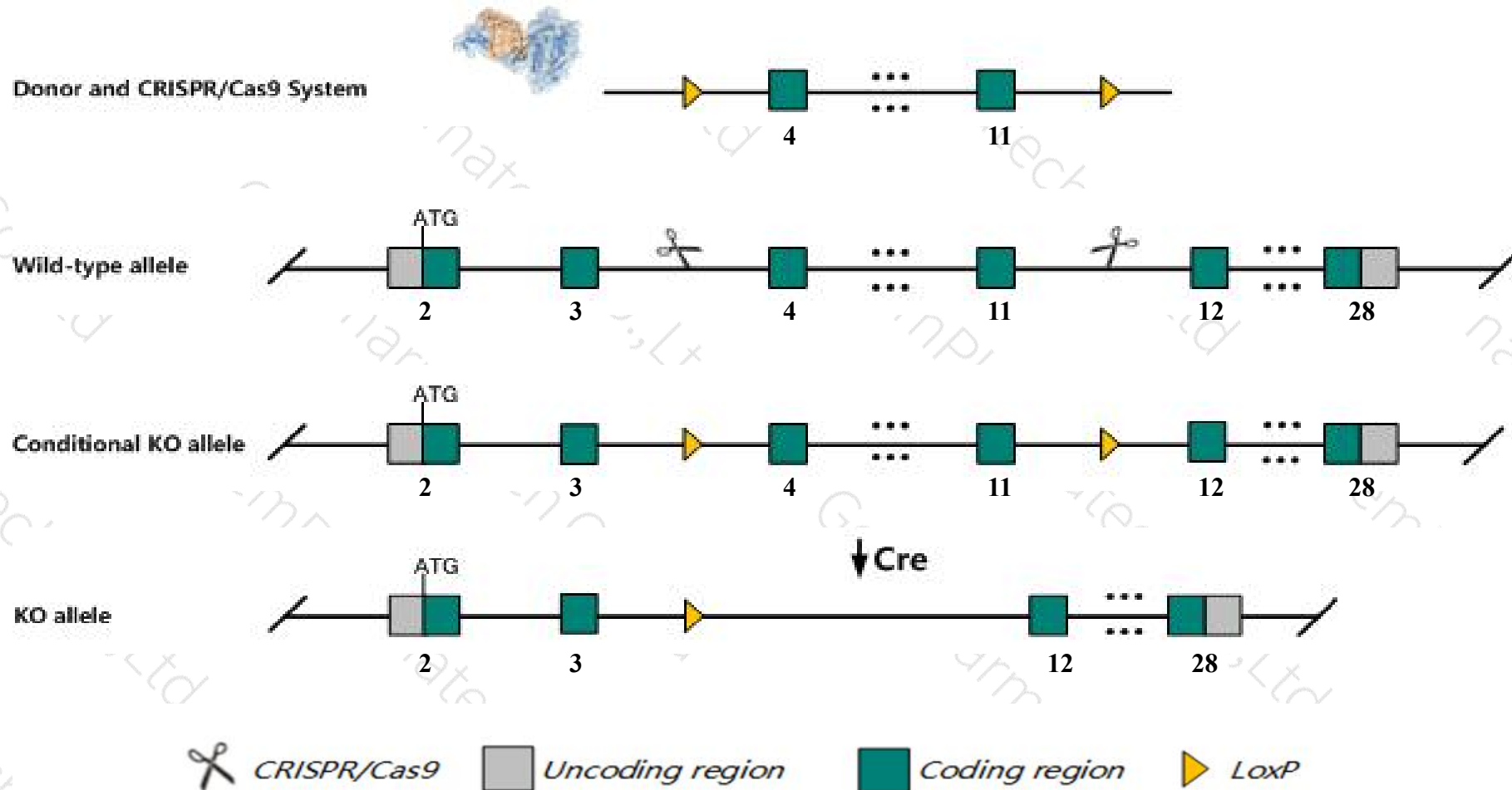
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Pfas* gene has 7 transcripts. According to the structure of *Pfas* gene, exon4-exon11 of *Pfas-201* (ENSMUST00000021282.11) transcript is recommended as the knockout region. The region contains 1058bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Pfas* 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, Mice heterozygous for spontaneous or ENU-induced mutations exhibit craniofacial abnormalities, most notably a domed cranium and short snout, variable white belly spots and white tail tips, and a range of eye defects including microphthalmia and anophthalmia.
- The *Pfas* gene is located on the Chr11. 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)

Pfas phosphoribosylformylglycinamide synthase (FGAR amidotransferase) [*Mus musculus* (house mouse)]

Gene ID: 237823, updated on 25-Feb-2020

Summary

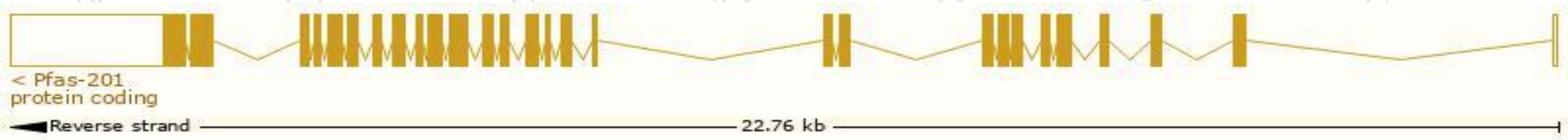
Official Symbol	Pfas provided by MGI
Official Full Name	phosphoribosylformylglycinamide synthase (FGAR amidotransferase) provided by MGI
Primary source	MGI:MGI:2684864
See related	Ensembl:ENSMUSG00000020899
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gm18; PURL; Sofa; FGAMS; FGARAT; FGAR-AT; 4432409B16Rik
Expression	Ubiquitous expression in CNS E11.5 (RPKM 10.9), ovary adult (RPKM 10.3) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

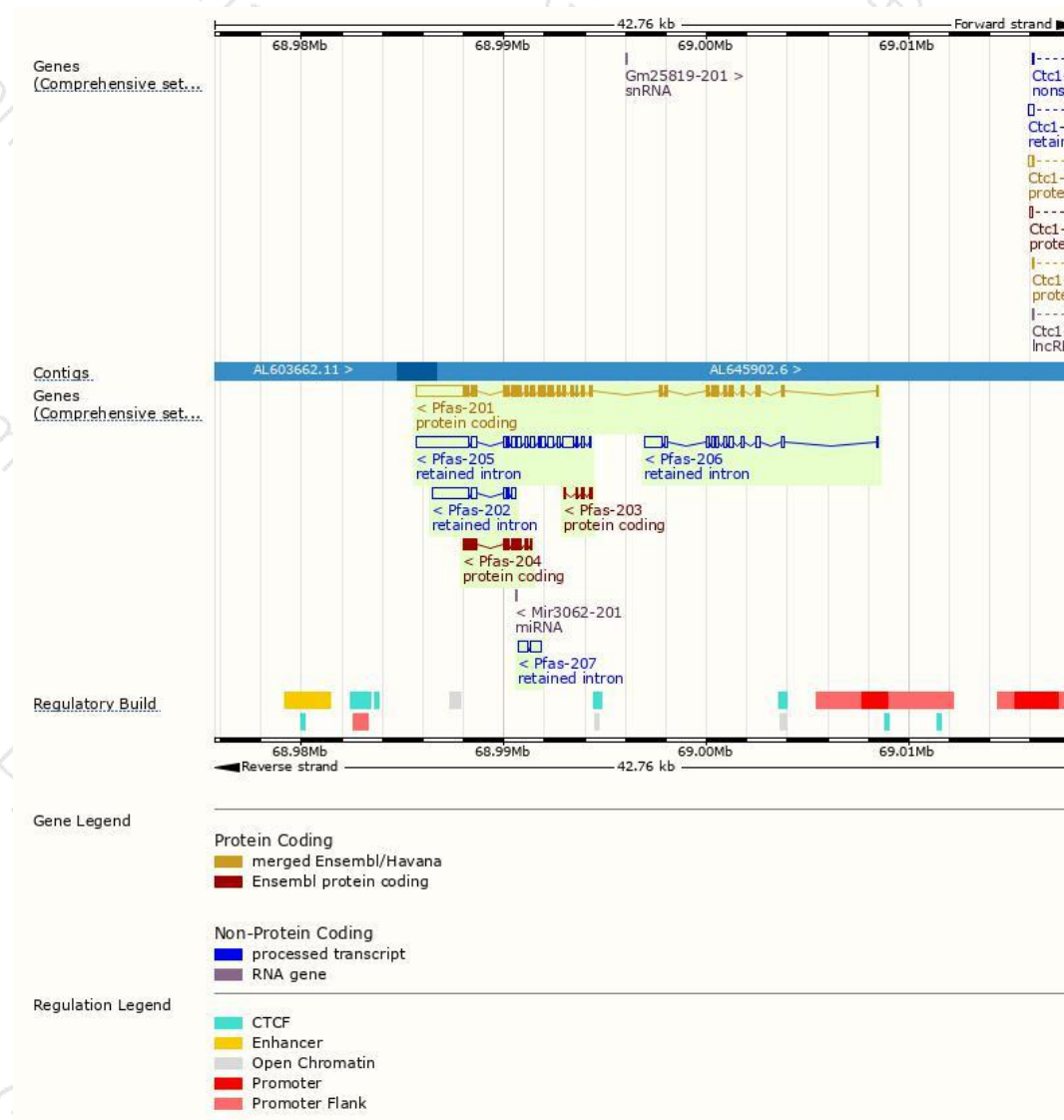
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pfas-201	ENSMUST00000021282.11	6373	1337aa	Protein coding	CCDS48823	Q5SUR0	TSL:5 GENCODE basic APPRIS P1
Pfas-204	ENSMUST00000152964.2	1421	473aa	Protein coding	-	F6RUL9	CDS 5' incomplete TSL:3
Pfas-203	ENSMUST00000149703.3	350	117aa	Protein coding	-	G3UY86	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Pfas-205	ENSMUST00000172915.7	5144	No protein	Retained intron	-	-	TSL:5
Pfas-202	ENSMUST00000146490.1	2584	No protein	Retained intron	-	-	TSL:1
Pfas-206	ENSMUST00000172987.1	2129	No protein	Retained intron	-	-	TSL:1
Pfas-207	ENSMUST00000173410.1	927	No protein	Retained intron	-	-	TSL:2

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



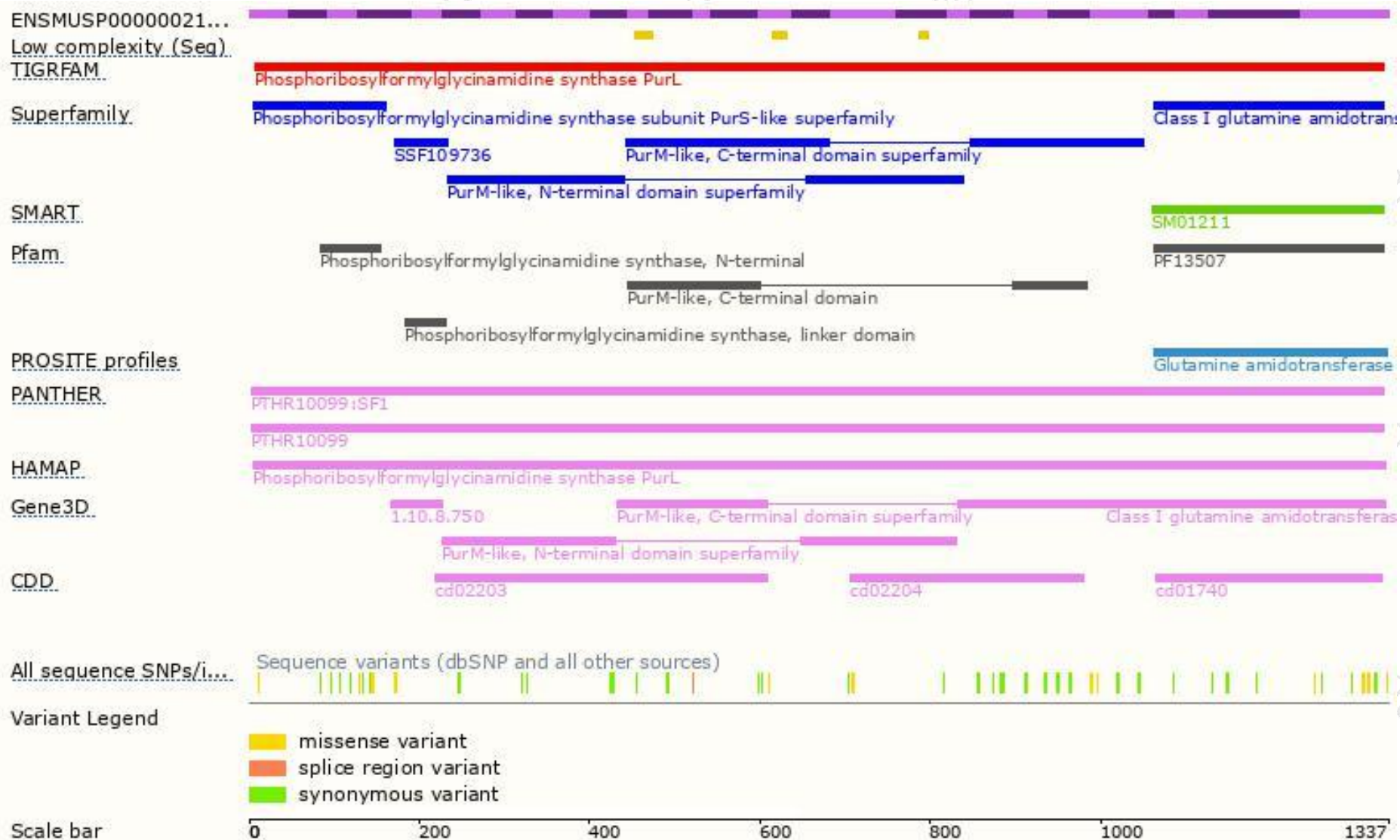
Genomic location distribution



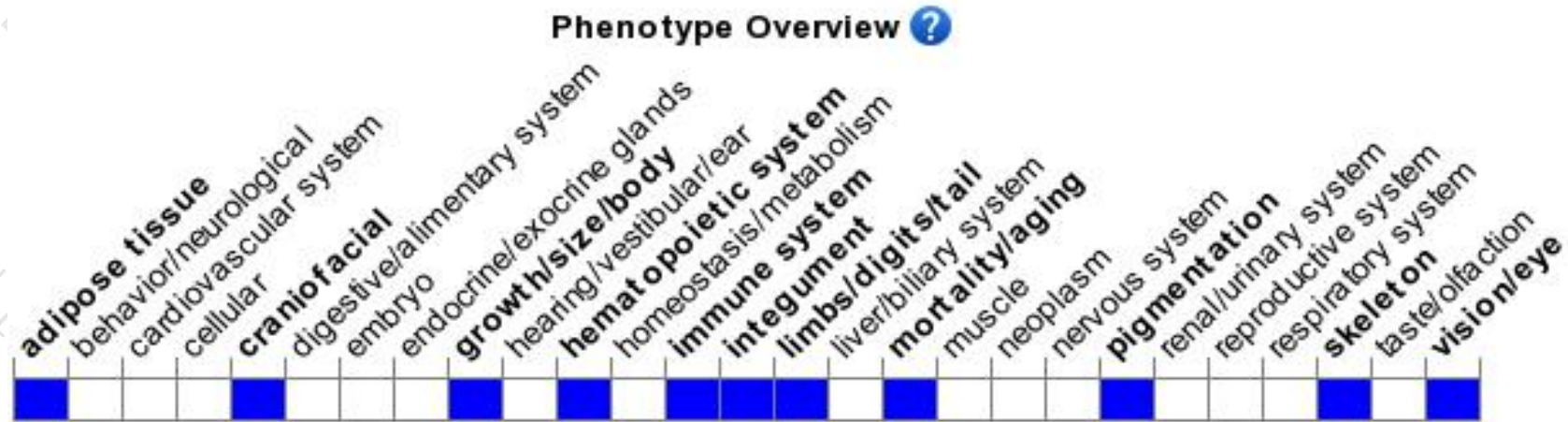
Protein domain



集萃药康
GemPharmatech



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 heterozygous for spontaneous or ENU-induced mutations exhibit craniofacial abnormalities, most notably a domed cranium and short snout, variable white belly spots and white tail tips, and a range of eye defects including microphthalmia and anophthalmia.

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

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