Selenof Cas9-CKO Strategy

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Reviewer: **Ruirui Zhang**

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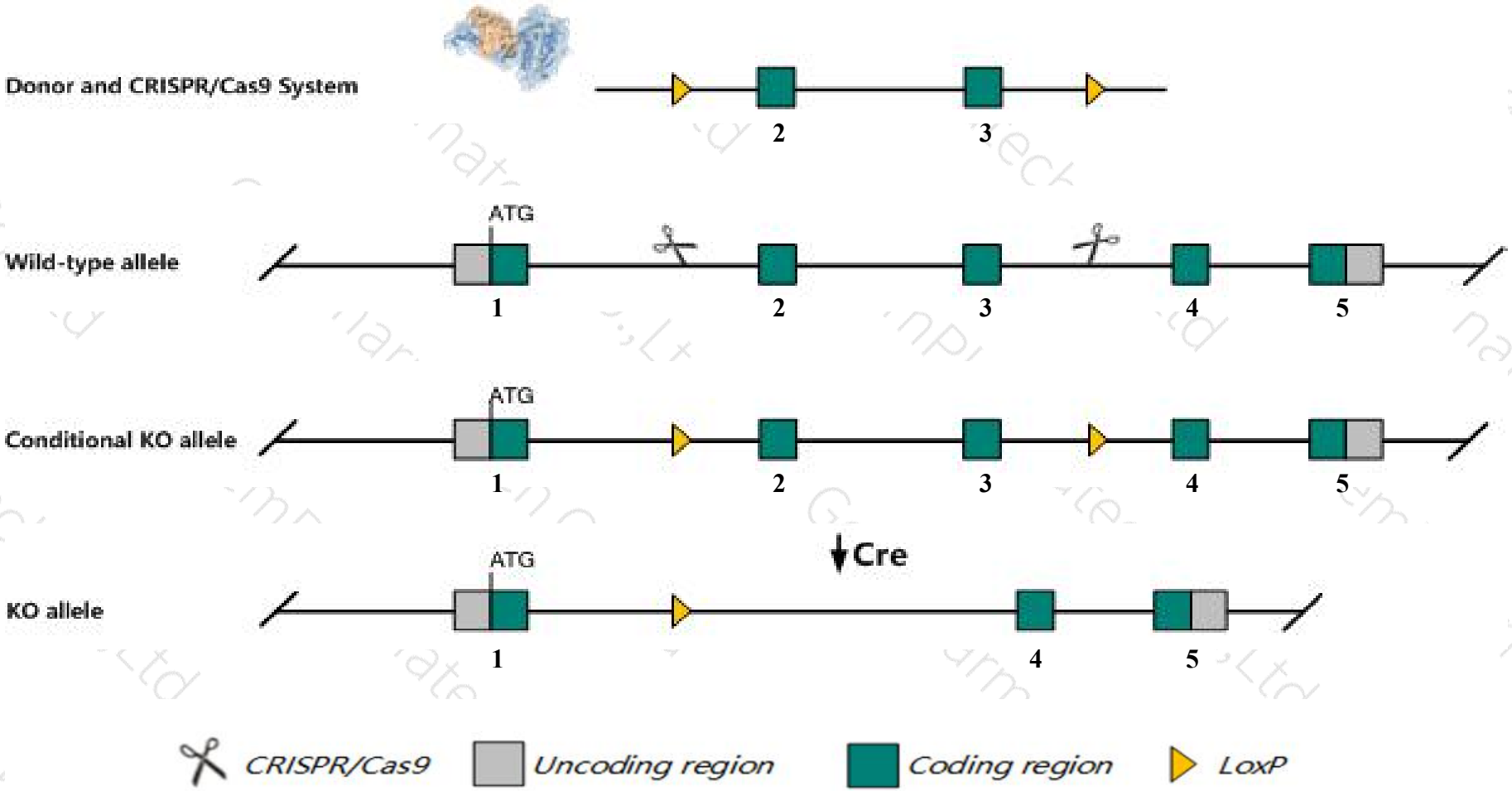
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Selenof* gene has 6 transcripts. According to the structure of *Selenof* gene, exon2-exon3 of *Selenof-201* (ENSMUST00000082437.9) transcript is recommended as the knockout region.The region contains 232bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Selenof* 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,Mice homozygous for a knock-out allele exhibit mild oxidative stress in the liver and develop cataracts by 1.5 months of age.

➢ The knockout region contains *Gm24406* gene.

➢ The *Selenof* gene is located on the Chr3. 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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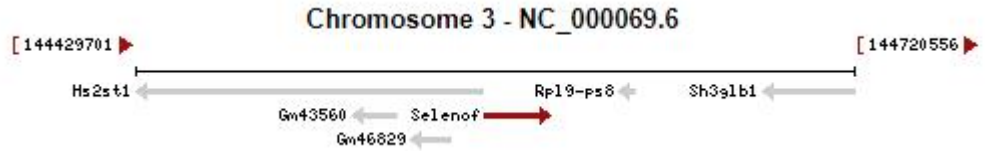
Selenof selenoprotein F [*Mus musculus* (house mouse)]

Gene ID: 93684, updated on 12-Nov-2019

Summary

Official Symbol	Selenof provided by MGI
Official Full Name	selenoprotein F provided by MGI
Primary source	MGI:MGI:1927947
See related	Ensembl:ENSMUSG00000037072
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Sep15; 9430015P09Rik
Summary	The protein encoded by this gene belongs to the SEP15/selenoprotein M family. The exact function of this protein is not known; however, it has been found to associate with UDP-glucose:glycoprotein glucosyltransferase (UGTR), an endoplasmic reticulum(ER)-resident protein, which is involved in the quality control of protein folding. The association with UGTR retains this protein in the ER, where it may play a role in protein folding. Knockout studies in mice also suggest a role for this gene in cataract formation and colon carcinogenesis. This protein is a selenoprotein, containing the rare amino acid selenocysteine (Sec). Sec is encoded by the UGA codon, which normally signals translation termination. The 3' UTRs of selenoprotein mRNAs contain a conserved stem-loop structure, designated the Sec insertion sequence (SECIS) element, that is necessary for the recognition of UGA as a Sec codon, rather than as a stop signal. [provided by RefSeq, Nov 2016]
Expression	Ubiquitous expression in placenta adult (RPKM 136.8), bladder adult (RPKM 131.5) and 28 other tissues See more
Orthologs	human all



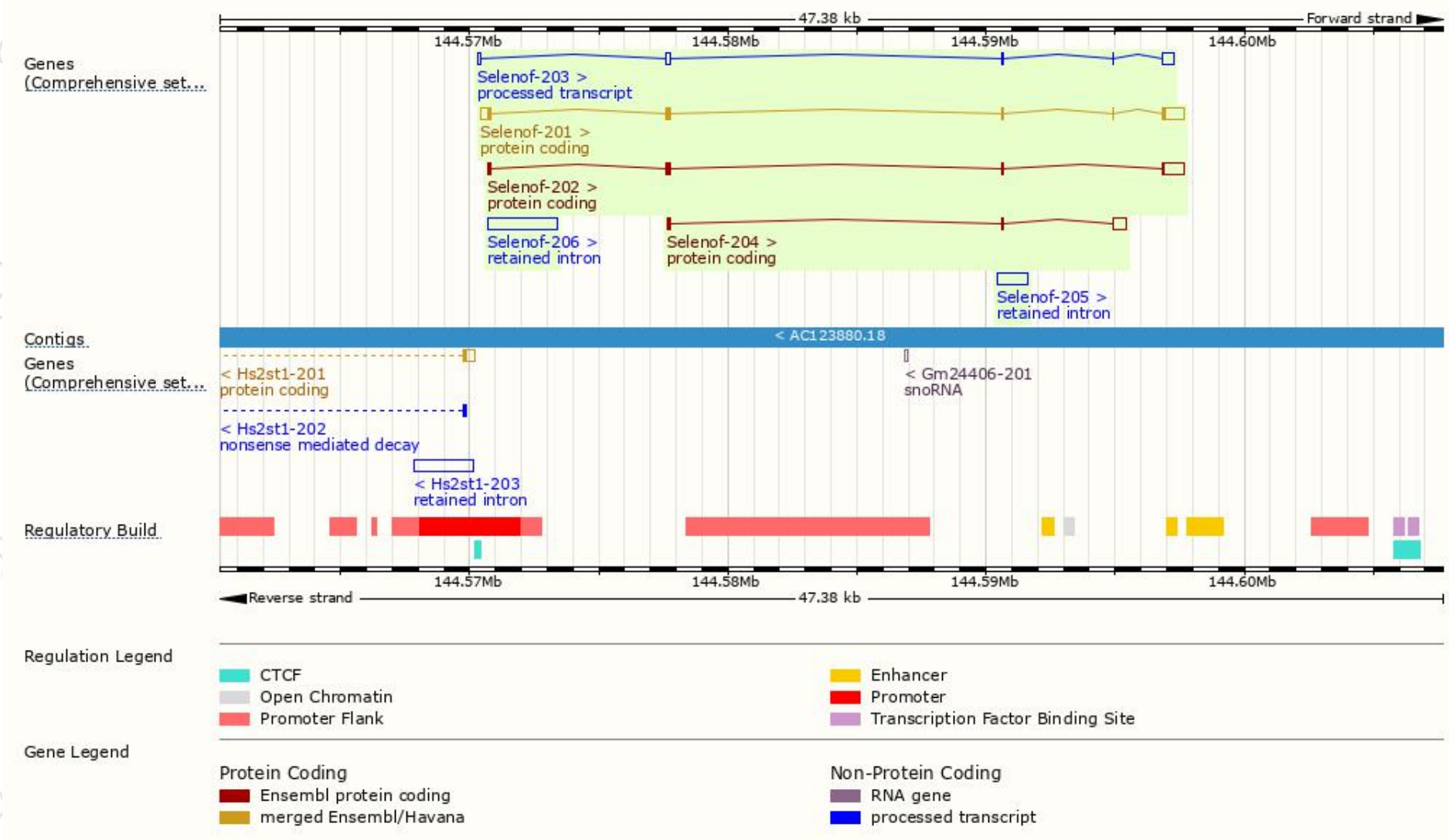
Transcript information（Ensembl）

The gene has 6 transcripts,all transcripts are shown below:

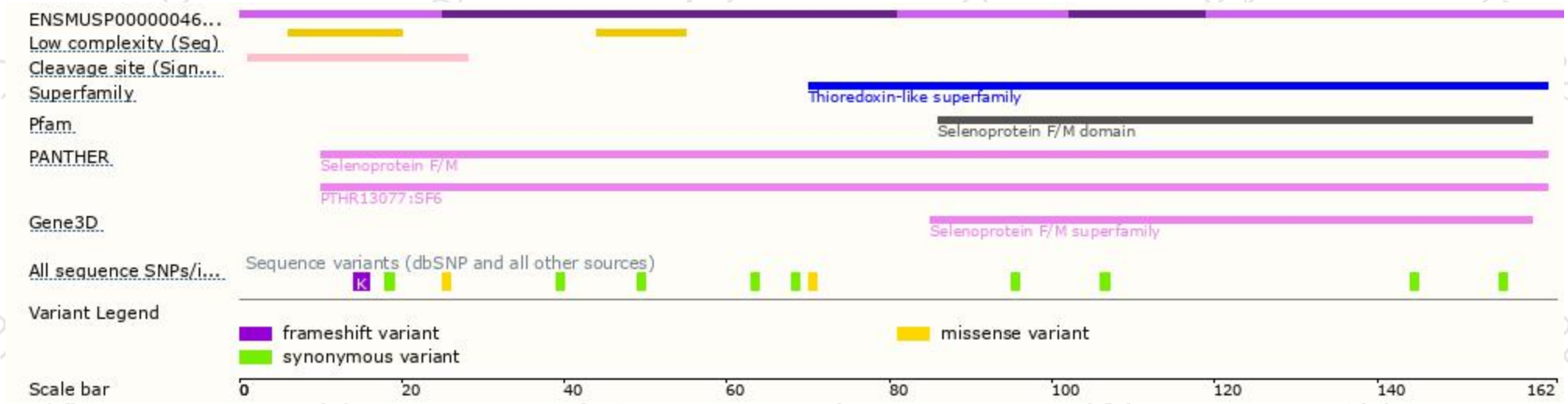
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Selenof-201	ENSMUST00000082437.9	1514	162aa	Protein coding	CCDS17884	A0A0R4J0K1	TSL:1 GENCODE basic APPRIS P1
Selenof-202	ENSMUST00000106211.1	1178	121aa	Protein coding	-	A0A1C7ZMY4	TSL:3 GENCODE basic
Selenof-204	ENSMUST00000151086.2	698	45aa	Protein coding	-	A0A0G2JEF3	CDS 5' incomplete TSL:2
Selenof-203	ENSMUST00000144859.3	825	No protein	Processed transcript	-	-	TSL:3
Selenof-206	ENSMUST00000198936.1	2699	No protein	Retained intron	-	-	TSL:NA
Selenof-205	ENSMUST00000198279.1	1166	No protein	Retained intron	-	-	TSL:NA

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

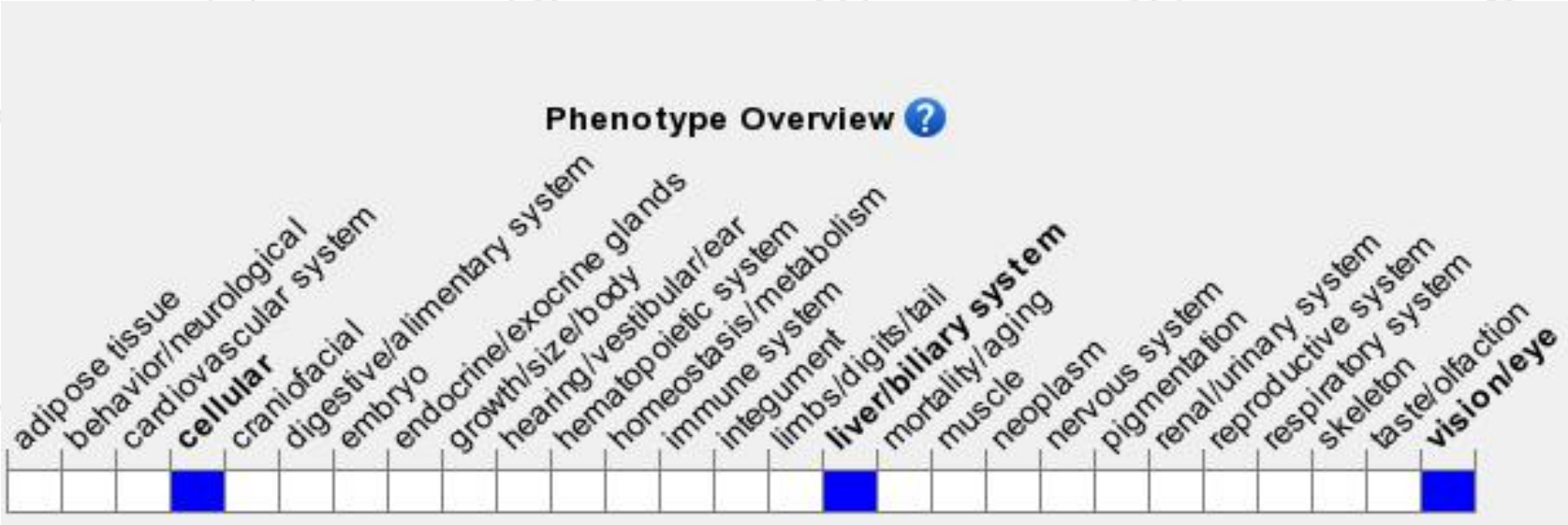
Genomic location distribution



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 mild oxidative stress in the liver and develop cataracts by 1.5 months of age.

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

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