

Usp8 Cas9-CKO Strategy

Designer:

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Design Date:

2018/5/31

Project Overview

Project Name

Usp8

Project type

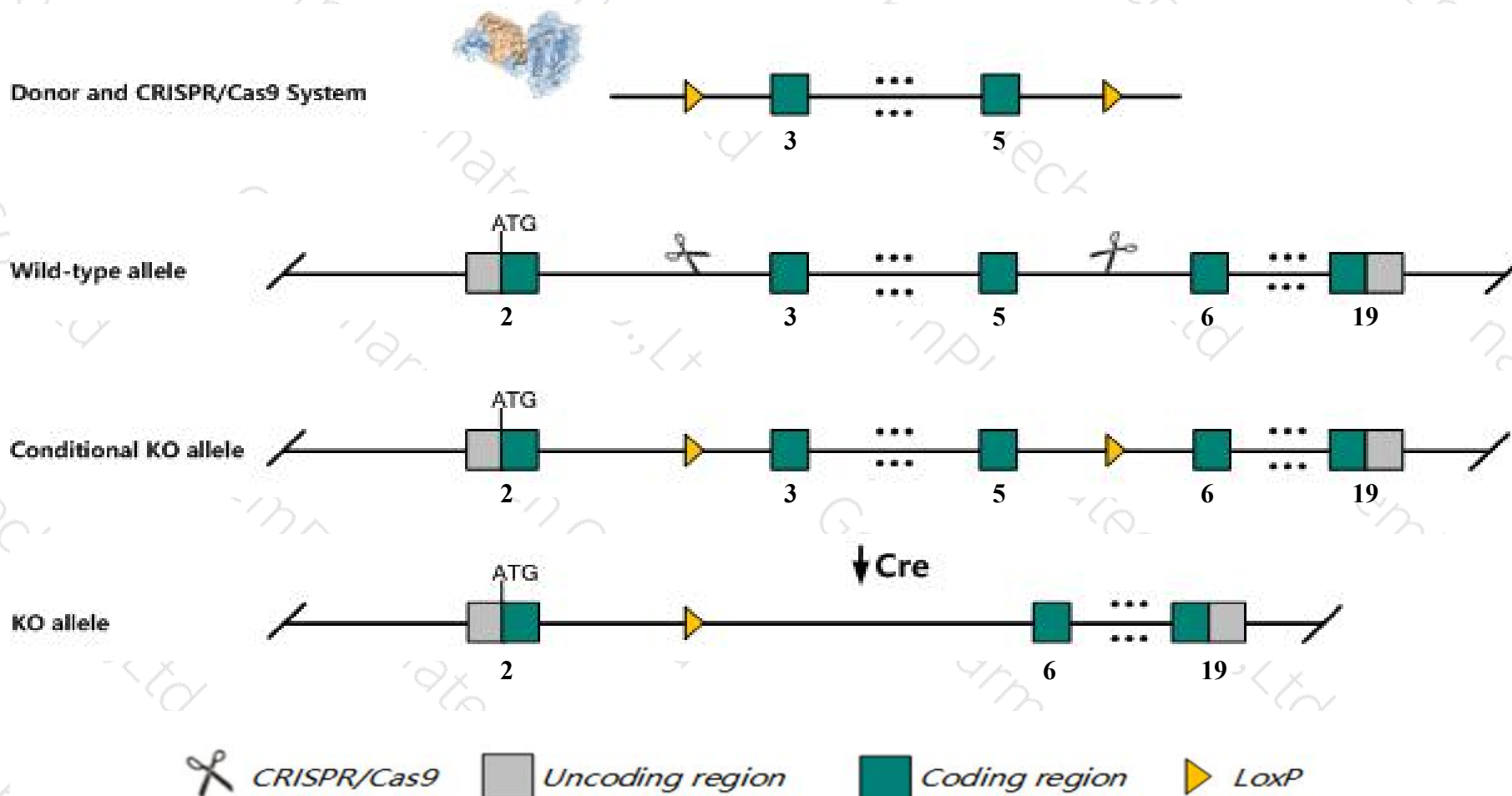
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Usp8* gene. The schematic diagram is as follows:



- The *Usp8* gene has 3 transcripts. According to the structure of *Usp8* gene, exon3-exon5 of *Usp8-202* (ENSMUST00000110416.2) transcript is recommended as the knockout region. The region contains 427bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Usp8* 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, Embryos homozygous for a knock-out allele appear severely growth retarded and disorganized at E9.5 and die in utero due to ventral-folding defects.
- The knockout region is located in the intron 7-8 of *Usp50-203*, and the effect on *Usp50-203* is unknown.
- The *Usp8* 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)

Usp8 ubiquitin specific peptidase 8 [Mus musculus (house mouse)]

Gene ID: 84092, updated on 5-Mar-2019

Summary



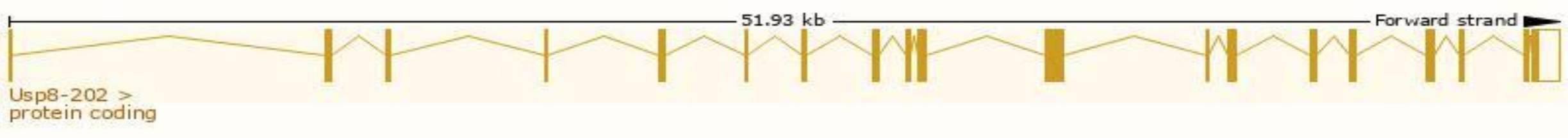
Official Symbol	Usp8 provided by MGI
Official Full Name	ubiquitin specific peptidase 8 provided by MGI
Primary source	MGI:MGI:1934029
See related	Ensembl:ENSMUSG00000027363
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI574262, AW557536, Ubpy, mKIAA0055
Expression	Ubiquitous expression in testis adult (RPKM 45.2), bladder adult (RPKM 14.4) and 25 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

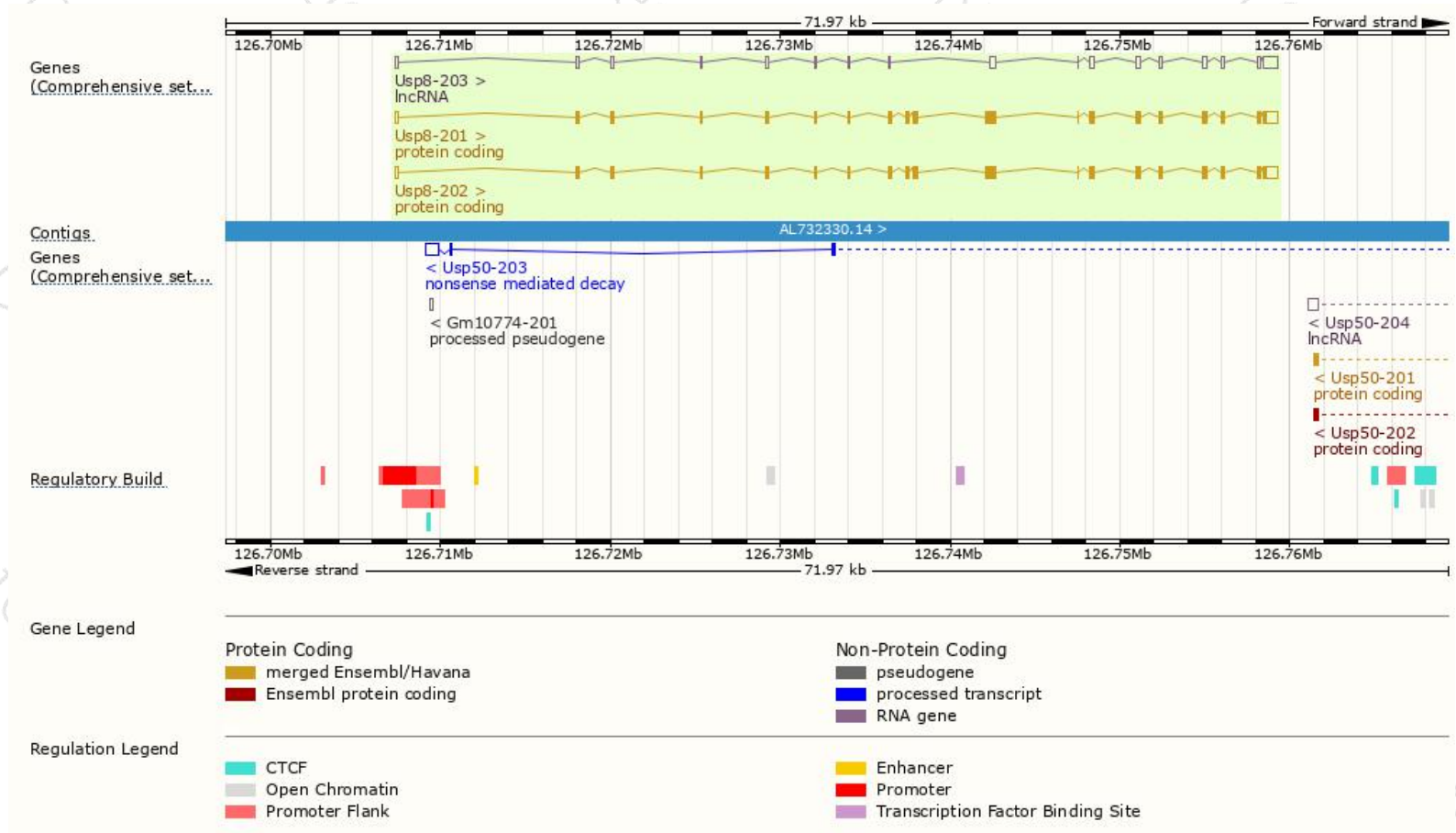
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Usp8-202	ENSMUST00000110416.2	4154	1091aa	Protein coding	CCDS57185	A2AI52	TSL:1 GENCODE basic APPRIS ALT2
Usp8-201	ENSMUST00000028841.13	4145	1080aa	Protein coding	CCDS16687	Q80U87	TSL:1 GENCODE basic APPRIS P3
Usp8-203	ENSMUST00000138859.1	3463	No protein	lncRNA	-	-	TSL:5

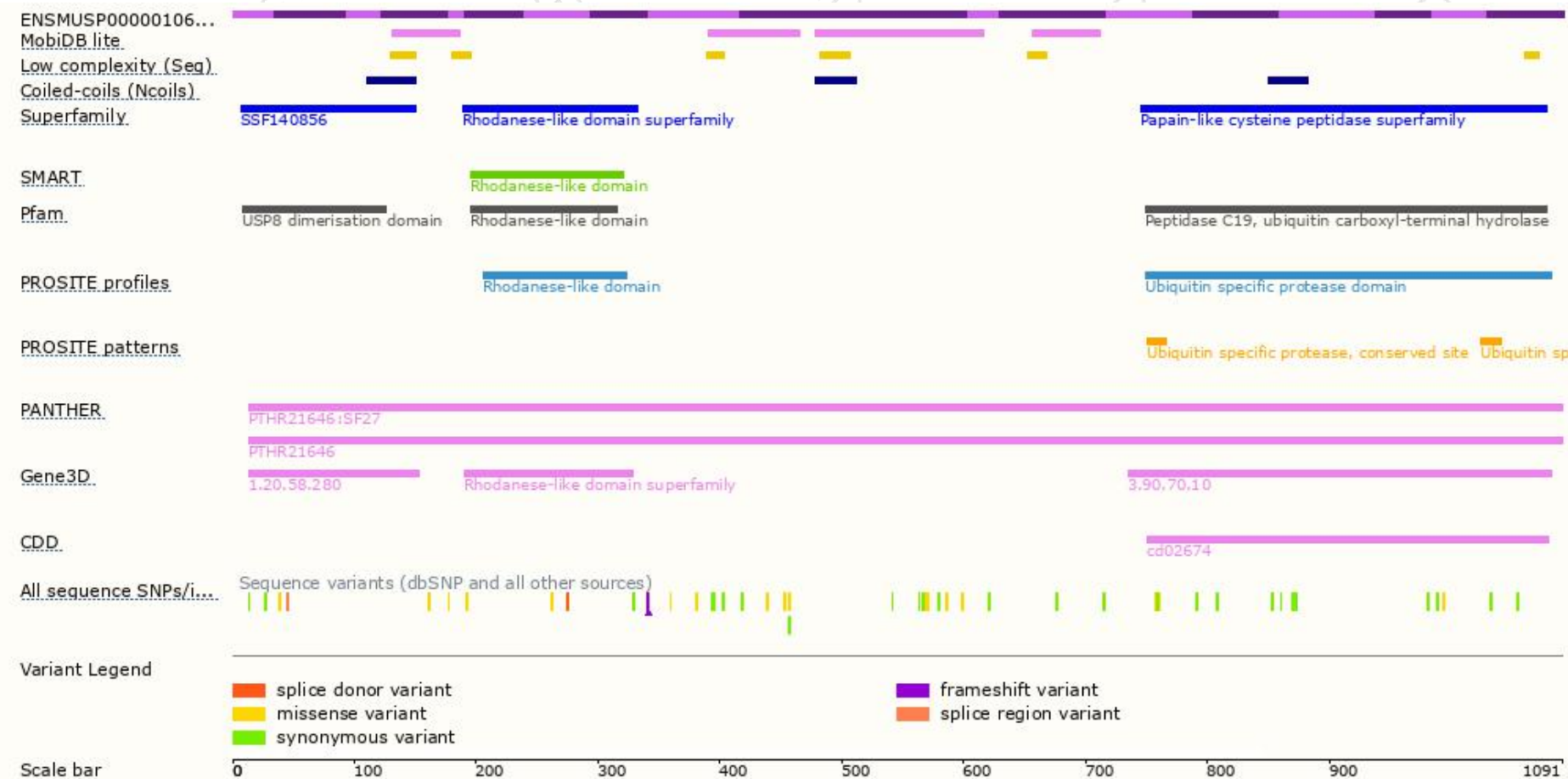
The strategy is based on the design of *Usp8-202* transcript,The transcription is shown below



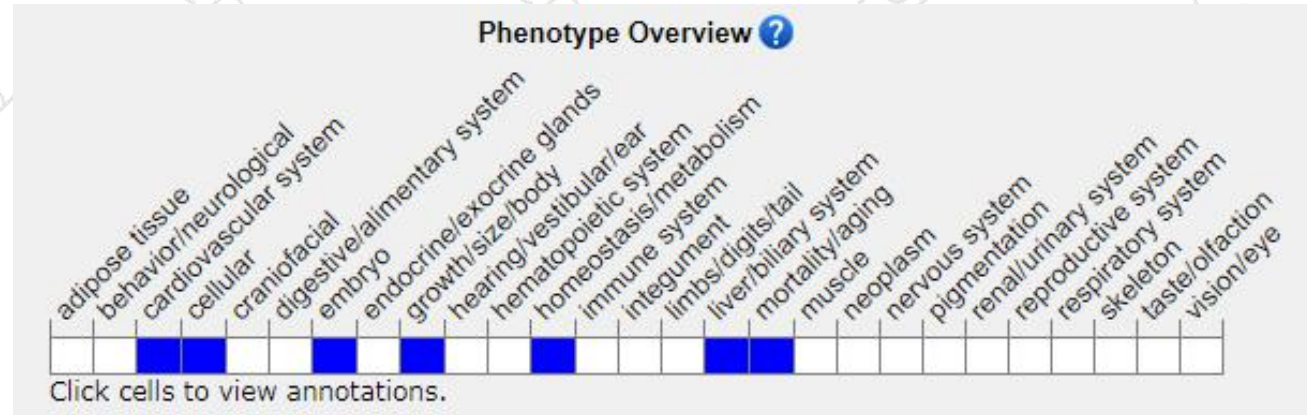
Genomic location distribution



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, Embryos homozygous for a knock-out allele appear severely growth retarded and disorganized at E9.5 and die in utero due to ventral-folding defects.

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

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