Sftpd Cas9-CKO Strategy

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Project Overview

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

Conditional Knockout strategy

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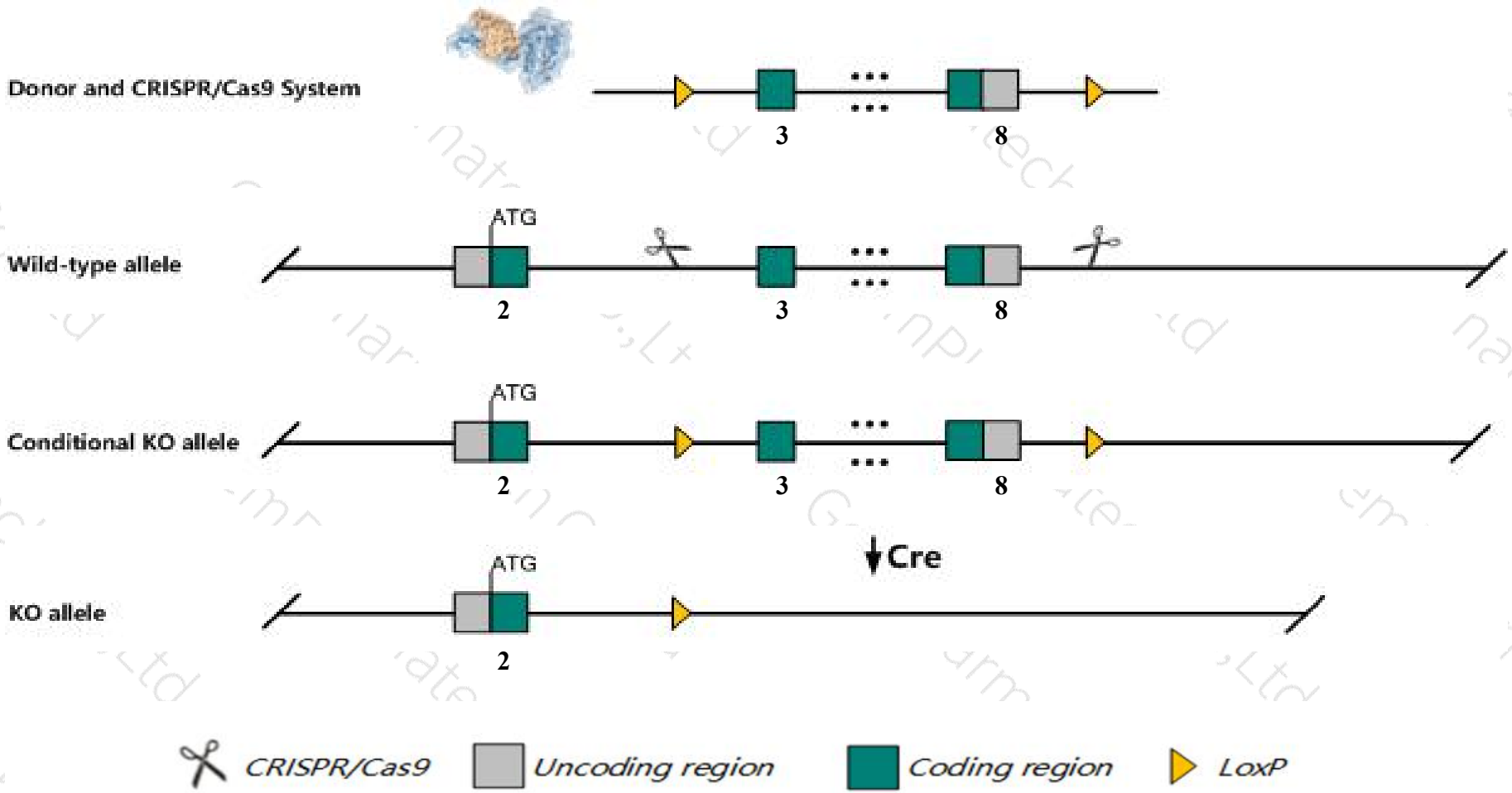
This model will use CRISPR/Cas9 technology to edit the *Sftpd* gene. The schematic diagram is as follows:



Technical routes

- The *Sftpd* gene has 2 transcripts. According to the structure of *Sftpd* gene, exon3-exon8 of *Sftpd-201* (ENSMUST00000077136.4) transcript is recommended as the knockout region.The region contains most of coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Sftpd* 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,Homozygotes for targeted null mutations exhibit increased pool sizes of alveolar and tissue phosphatidylcholine, accumulation of surfactant lipids, altered phospholipid structure, emphysema, and pulmonary fibrosis and chronic inflammation.

- The *Sftpd* gene is located on the Chr14. 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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Sftpd surfactant associated protein D [*Mus musculus* (house mouse)]

Gene ID: 20390, updated on 12-Nov-2019

Summary

Official Symbol	Sftpd provided by MGI
Official Full Name	surfactant associated protein D provided by MGI
Primary source	MGI:MGI:109515
See related	Ensembl:ENSMUSG00000021795
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	SP-D; Sfpd; Sftp4; AI573415
Expression	Restricted expression toward lung adult (RPKM 105.6) See more
Orthologs	human all

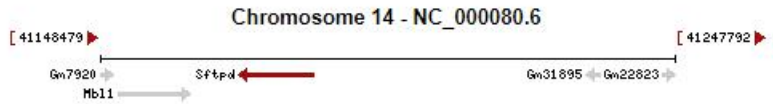
Genomic context

Location: 14 B; 14 22.36 cM

See Sftpd in Genome Data Viewer

Exon count: 8

Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	14	NC_000080.6 (41172212..41185198, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	14	NC_000080.5 (41985501..41998487, complement)

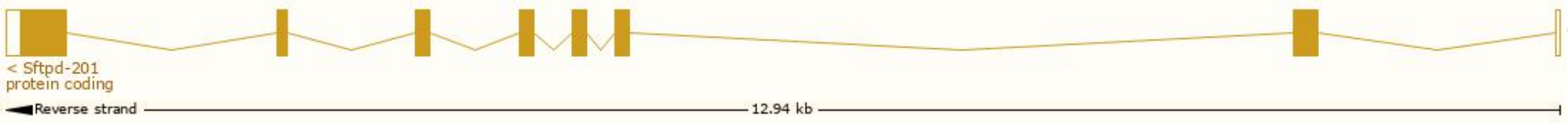


Transcript information（Ensembl）

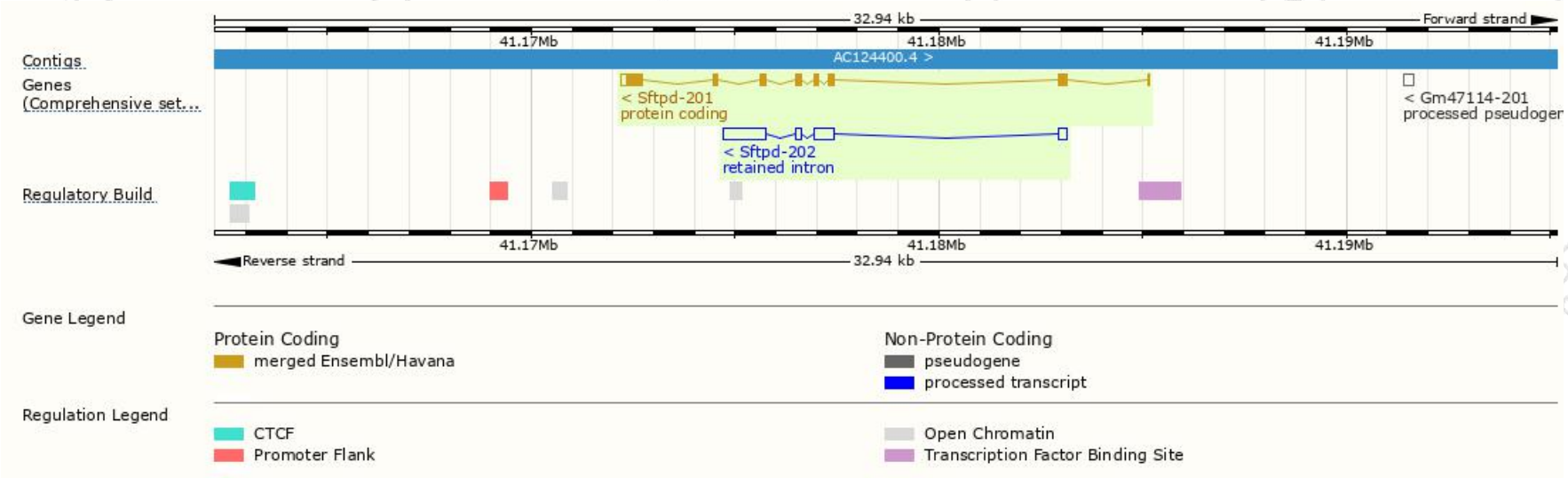
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sftpd-201	ENSMUST00000077136.4	1275	374aa	Protein coding	CCDS26962	P50404	TSL:1 GENCODE basic APPRIS P1
Sftpd-202	ENSMUST00000225892.1	1809	No protein	Retained intron	-	-	-

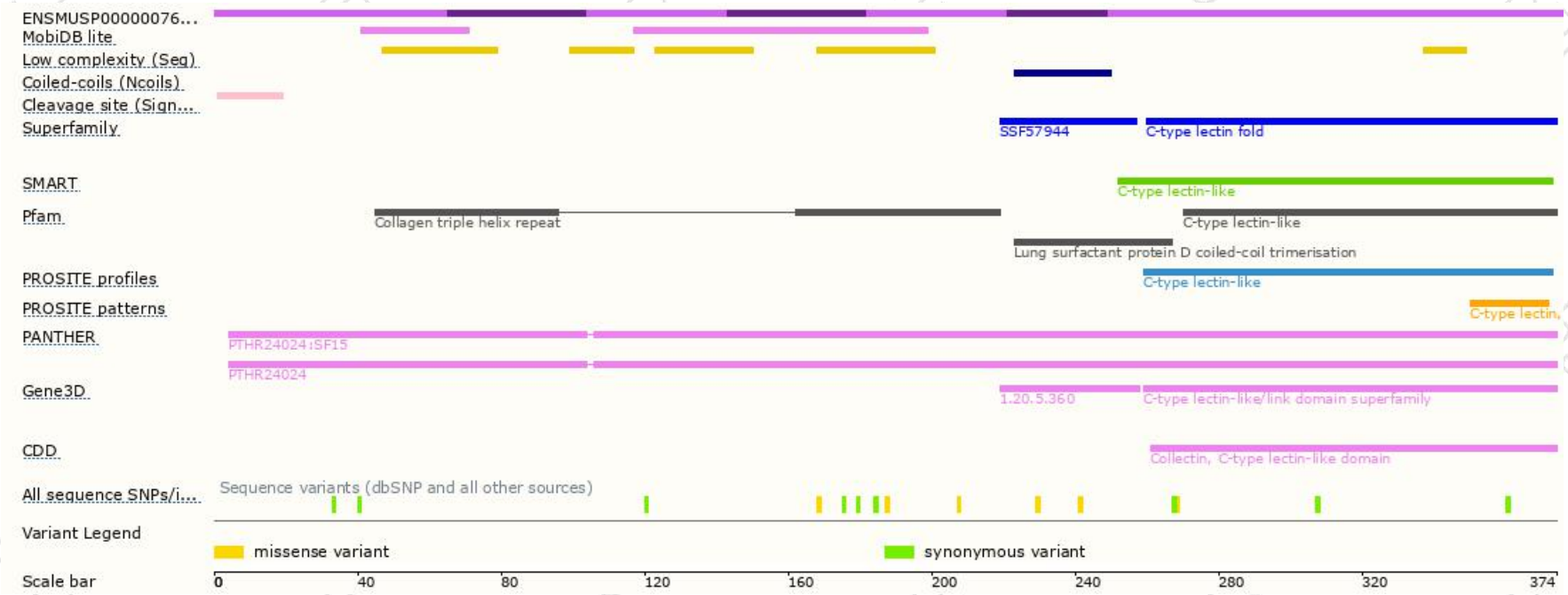
The strategy is based on the design of *Sftpd-201* transcript,The transcription is shown below



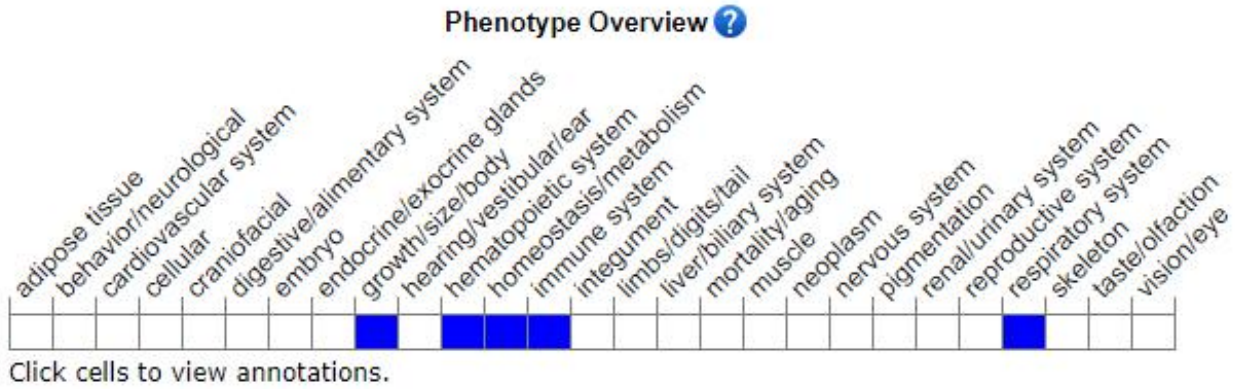
Genomic location distribution



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/).

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

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