Sox3 Cas9-CKO Strategy

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Project Overview

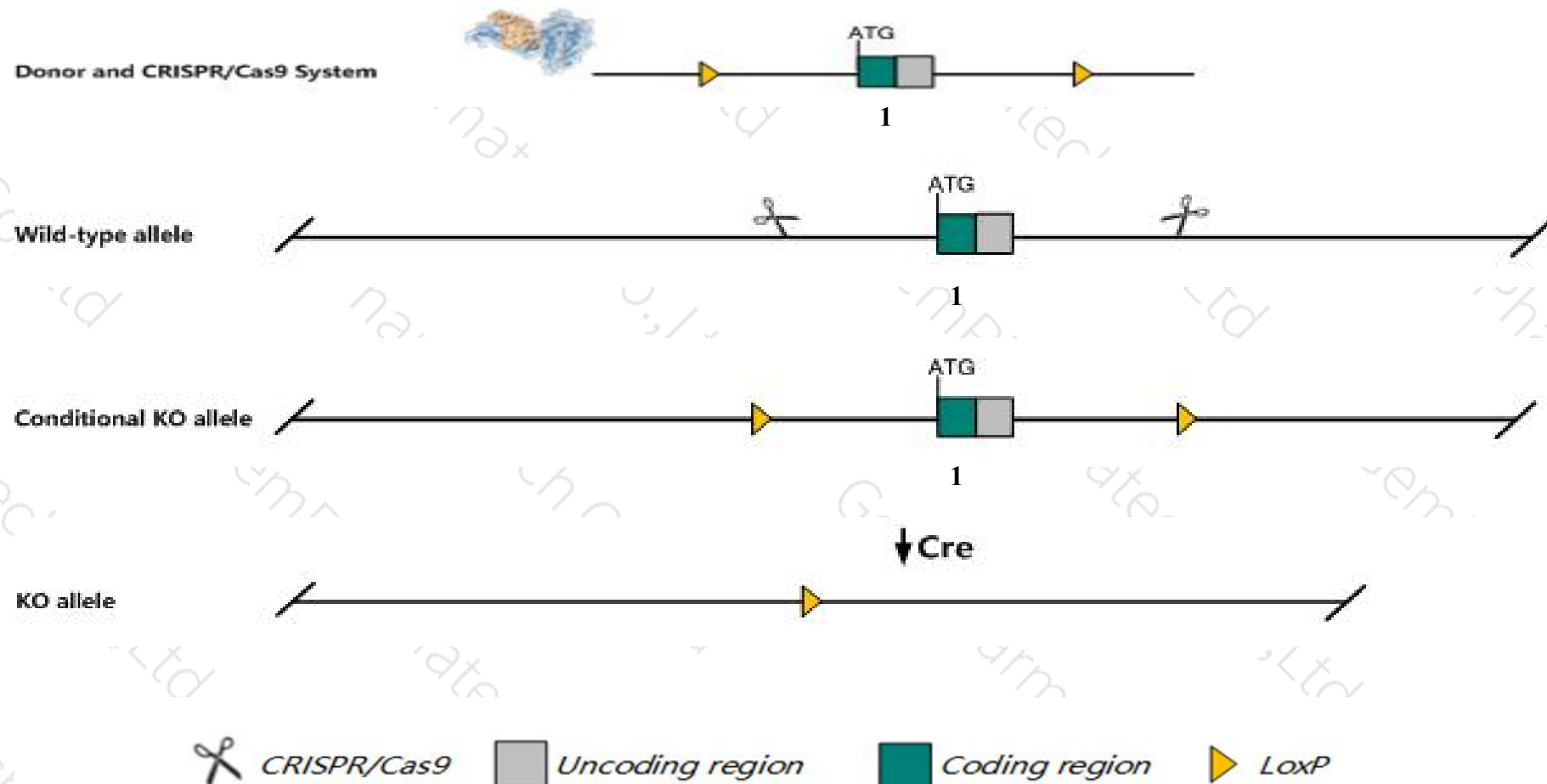|	
Project Name	*Sox3*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sox3* gene. The schematic diagram is as follows:

CRISPR/Cas9   Uncoding region   Coding region   LoxP

Technical routes

➢ The *Sox3* gene has 1 transcript. According to the structure of *Sox3* gene, exon1 of *Sox3-201* (ENSMUST00000135107.3) transcript is recommended as the knockout region.The region contains all 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 *Sox3* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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,Sex determination is normal in both homozygous mutant female and hemizygous mutant male mice, however, gonadal and developmental defects are observed in both sexes.

➢ The KO region contains part region of the *Gm14662* gene. The *Gm14662* gene will be deleted together.

➢ The *Sox3* gene is located on the ChrX. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information （NCBI）

Sox3 SRY (sex determining region Y)-box 3 [Mus musculus (house mouse)]

Gene ID: 20675, updated on 31-Jan-2019

Summary

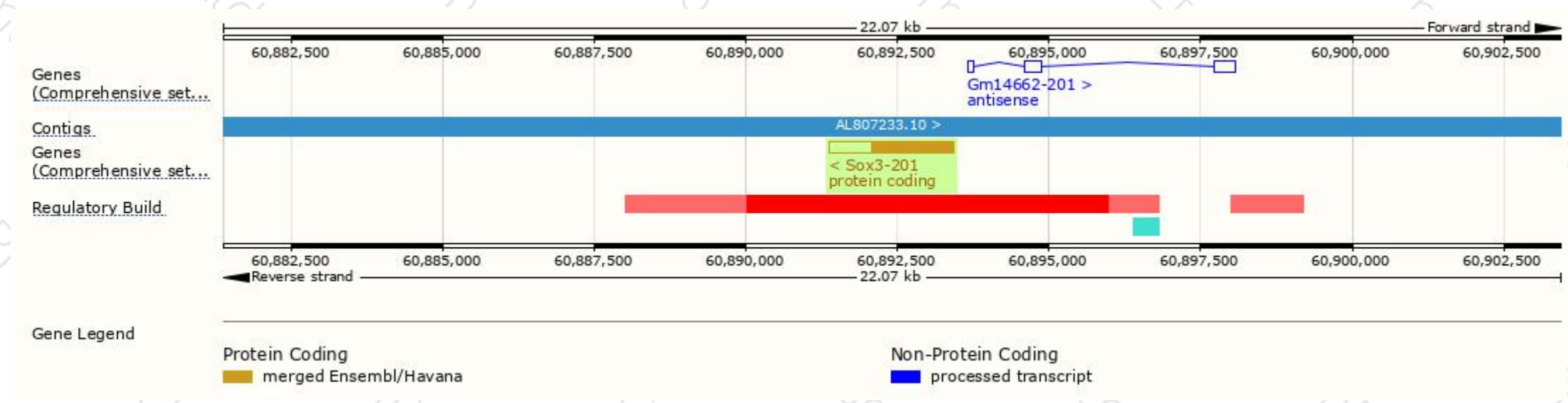|	
Official Symbol	Sox3 provided by MGI
Official Full Name	SRY (sex determining region Y)-box 3 provided by MGI
Primary source	MGI:MGI:98365
See related	Ensembl:ENSMUSG00000045179
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Sox-3
Orthologs	human all

Transcript information（Ensembl）

The gene has 1 transcript,and the transcript is shown below:

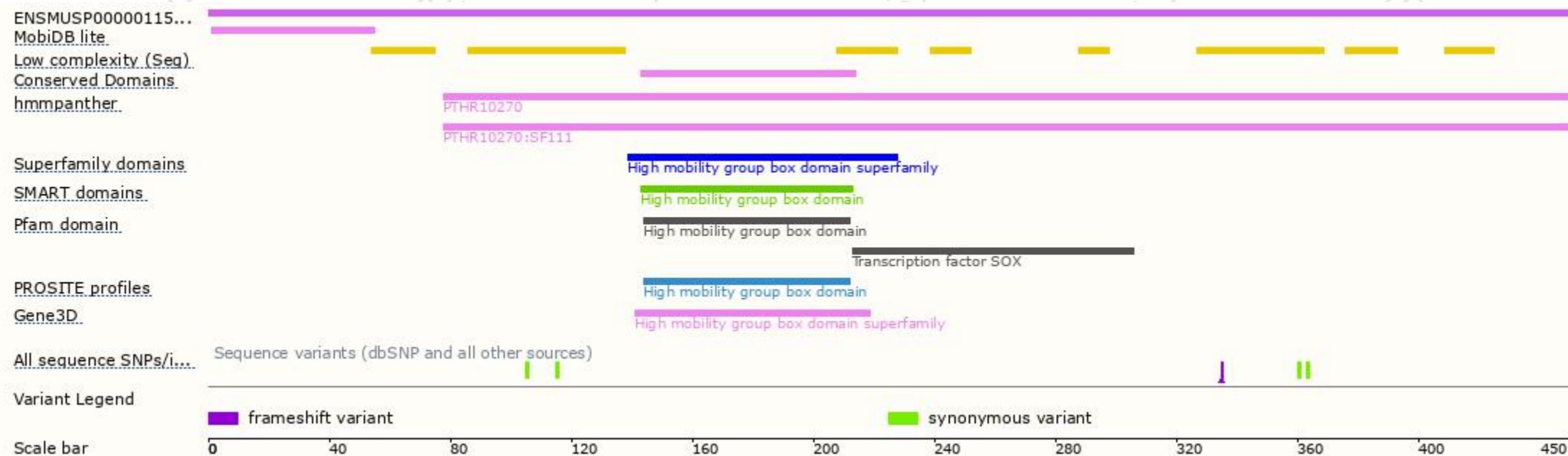| Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sox3-201	ENSMUST00000135107.3	2065	450aa	Protein coding	CCDS30161	A2AM37	TSL:NA GENCODE basic APPRIS P1

The strategy is based on the design of *Sox3-201* transcript,The transcription is shown below
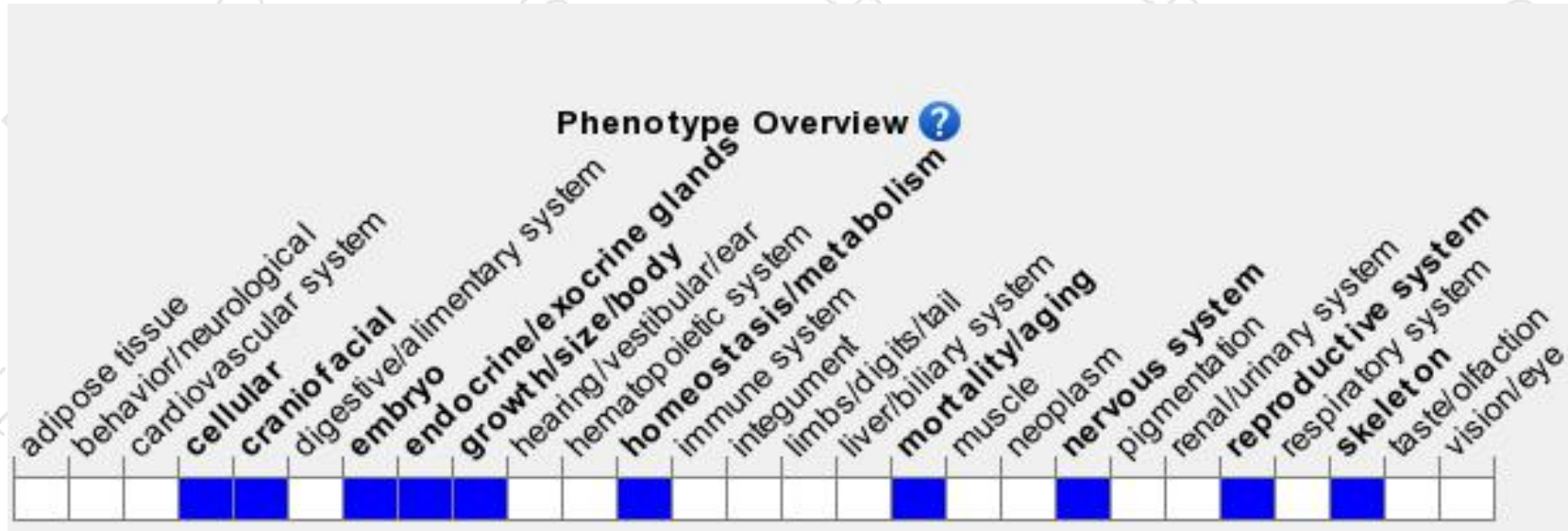

< Sox3-201
protein coding

Reverse strand   2.06 kb

Genomic location distribution

Protein domain

| Scale bar | 0 | 40 | 80 | 120 | 160 | 200 | 240 | 280 | 320 | 360 | 400 | 450 |

ENSMUSP00000115...
MobiDB lite
Low complexity (Seg)
Conserved Domains
hmmpanther
    PTHR10270
    PTHR10270:SF111
Superfamily domains   High mobility group box domain superfamily
SMART domains   High mobility group box domain
Pfam domain   High mobility group box domain
     Transcription factor SOX
PROSITE profiles   High mobility group box domain
Gene3D   High mobility group box domain superfamily
All sequence SNPs/i....   Sequence variants (dbSNP and all other sources)
Variant Legend   frameshift variant   synonymous variant

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,Sex determination is normal in both homozygous mutant female and hemizygous mutant male mice, however, gonadal and developmental defects are observed in both sexes.

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