Sympk Cas9-CKO Strategy

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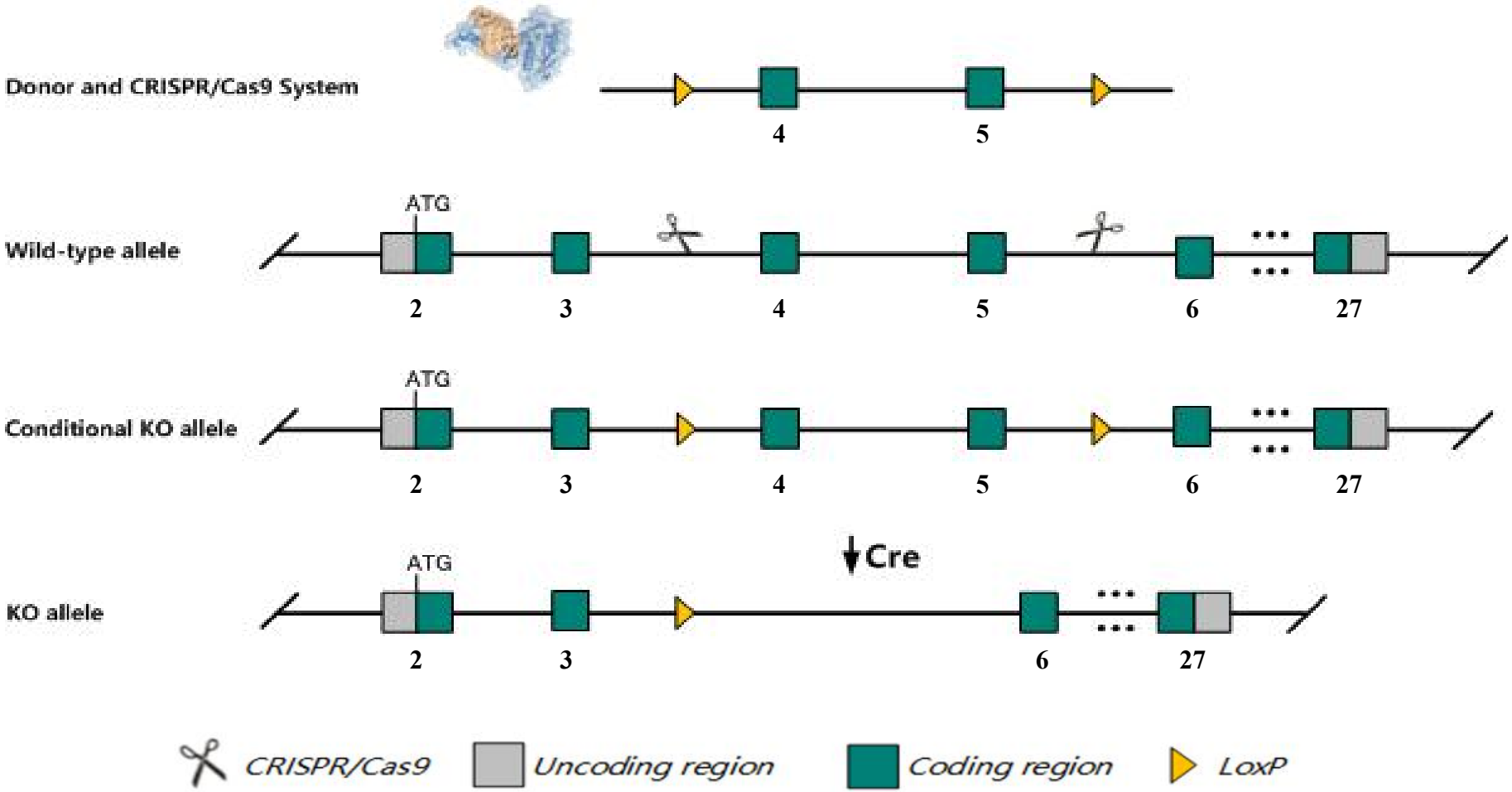
Project Overview

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

Conditional Knockout strategy

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



Technical routes

The *Sympk* gene has 8 transcripts. According to the structure of *Sympk* gene, exon4-exon5 of *Sympk-201*(ENSMUST00000023882.14) transcript is recommended as the knockout region.The region contains 128bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Sympk* 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

Transcript *Sympk-202* may not be affected.

According to the existing MGI data,mice homozygous ofr a transgenic gene disruption exhibit anemia at E15 and hydrops fetalis.

The *Sympk* gene is located on the Chr7. 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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Sympk symplekin [Mus musculus (house mouse)]

Gene ID: 68188, updated on 29-Jan-2021

Summary

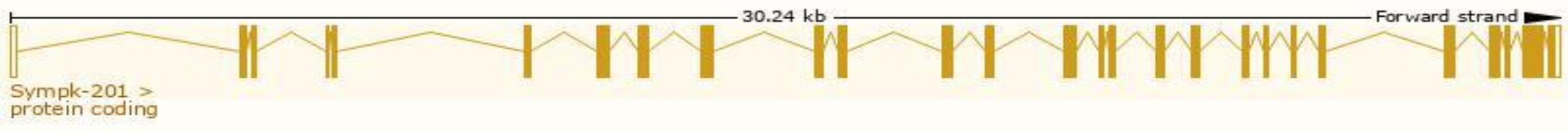
Official Symbol	Sympk provided by MGI
Official Full Name	symplekin provided byMGI
Primary source	MGI:MGI:1915438
See related	Ensembl:ENSMUSG00000023118
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1500016F02Rik, 4632415H16Rik, AA125406, AI449890, SPK, SYM
Expression	Ubiquitous expression in testis adult (RPKM 36.3), ovary adult (RPKM 34.7) and 28 other tissuesSee more
Orthologs	human all

Transcript information Ensembl

The gene has 8 transcripts,all transcripts are shown below:

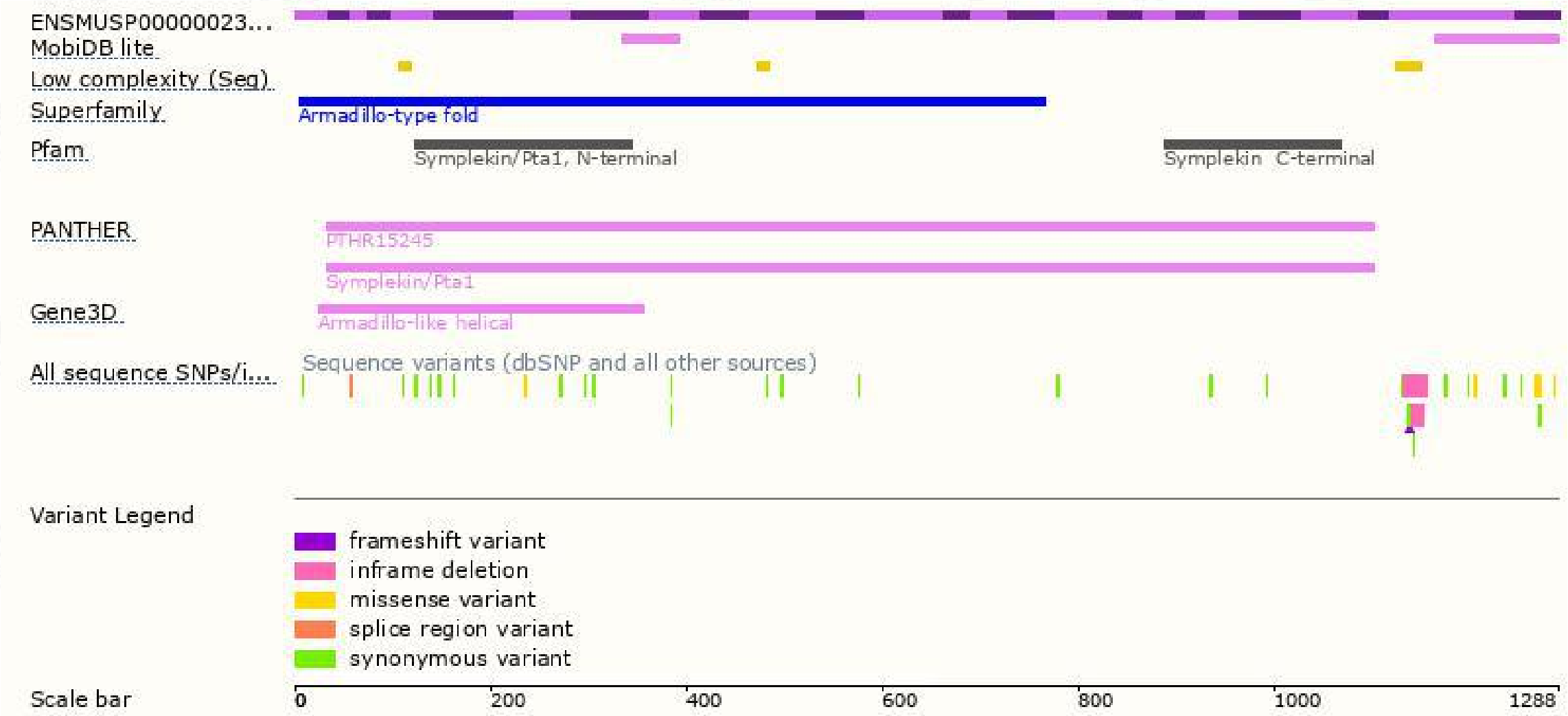
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sympk-201	ENSMUST00000023882.14	4116	1288aa	Protein coding	CCDS52051		TSL:1 , GENCODE basic , APPRIS P1 ,
Sympk-208	ENSMUST00000153976.8	827	198aa	Protein coding	-		CDS 3' incomplete , TSL:5 ,
Sympk-202	ENSMUST00000130328.2	469	143aa	Protein coding	-		CDS 5' incomplete , TSL:3 ,
Sympk-206	ENSMUST00000146903.8	4781	234aa	Nonsense mediated decay	-		TSL:1 ,
Sympk-207	ENSMUST00000148861.8	1099	No protein	Processed transcript	-		TSL:1 ,
Sympk-205	ENSMUST00000138440.2	799	No protein	Processed transcript	-		TSL:3 ,
Sympk-204	ENSMUST00000137287.3	4760	No protein	Retained intron	-		TSL:1 ,
Sympk-203	ENSMUST00000131230.2	636	No protein	Retained intron	-		TSL:2 ,

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

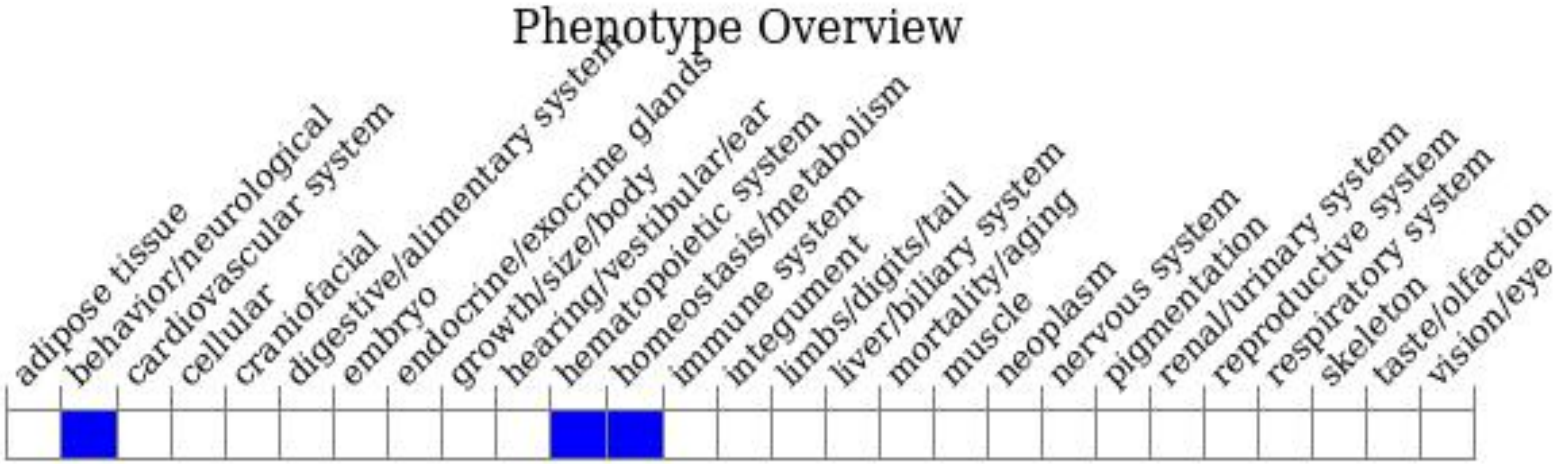


Genomic location distribution

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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