Shb Cas9-KO Strategy

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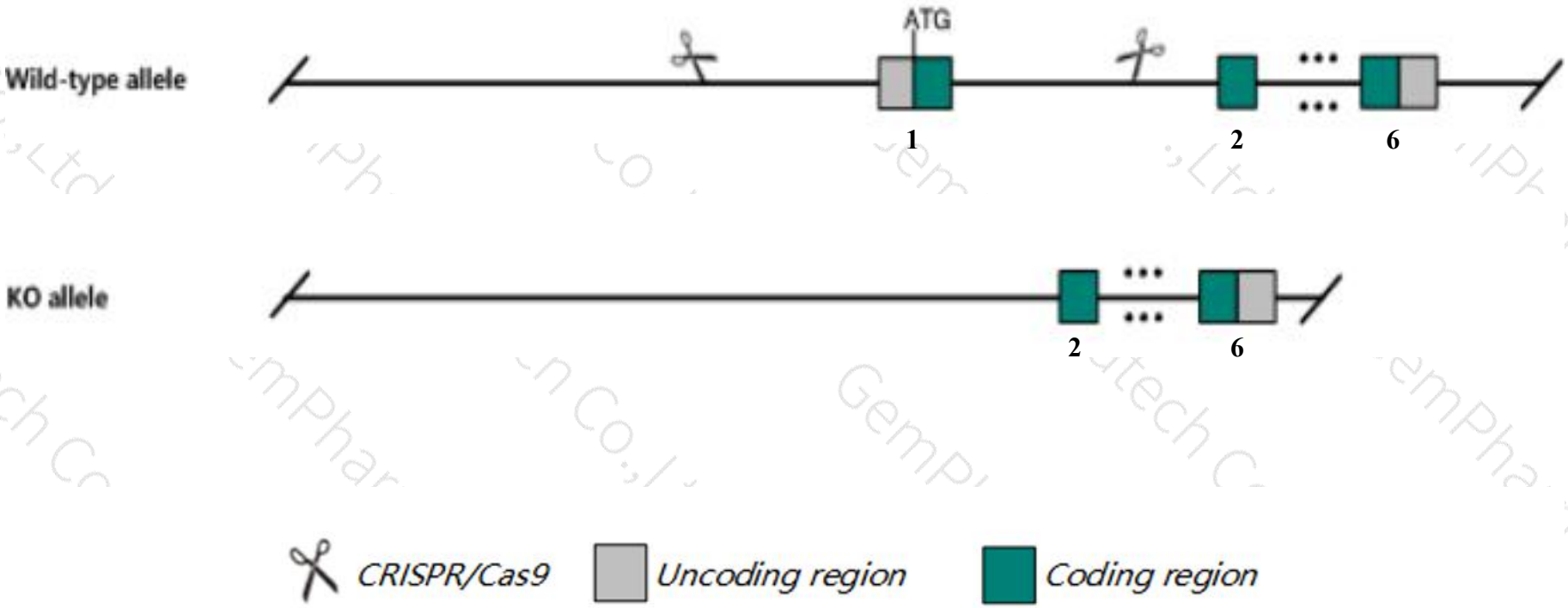
Project Overview

Project Name	***Shb***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

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



Technical routes

➢ The *Shb* gene has 6 transcripts. According to the structure of *Shb* gene, exon1 of *Shb-201*(ENSMUST00000061986.11) transcript is recommended as the knockout region.The region contains start codon ATG.Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Shb* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ According to the existing MGI data,mice homozygous for a null allele exhibit some embryonic lethality associated with various embryogenesis defects including loss of embryo structures, open neural tube, hemorrhaging and tail defects. Heterozygous mice exhibit a distortion in the transmission ratio of the allele maternally.

➢ The effect on transcript *Shb*-204 is unknown.

➢ Transcript *Shb*-202&203&206 may not be affected.

➢ The *Shb* gene is located on the Chr4. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

Shb src homology 2 domain-containing transforming protein B [Mus musculus (house mouse)]

Gene ID: 230126, updated on 13-Mar-2020

Summary

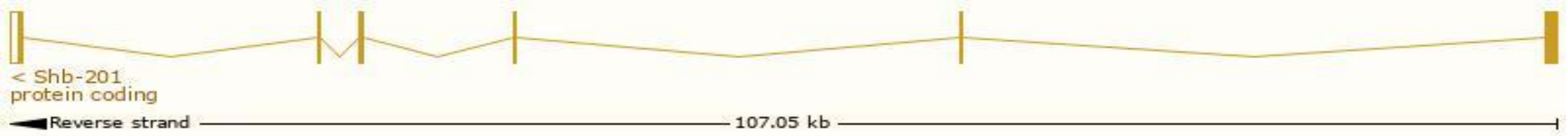
Official Symbol	Shb provided by MGI
Official Full Name	src homology 2 domain-containing transforming protein B provided byMGI
Primary source	MGI:MGI:98294
See related	Ensembl:ENSMUSG00000044813
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BC028832
Expression	Broad expression in adrenal adult (RPKM 54.6), duodenum adult (RPKM 22.8) and 25 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

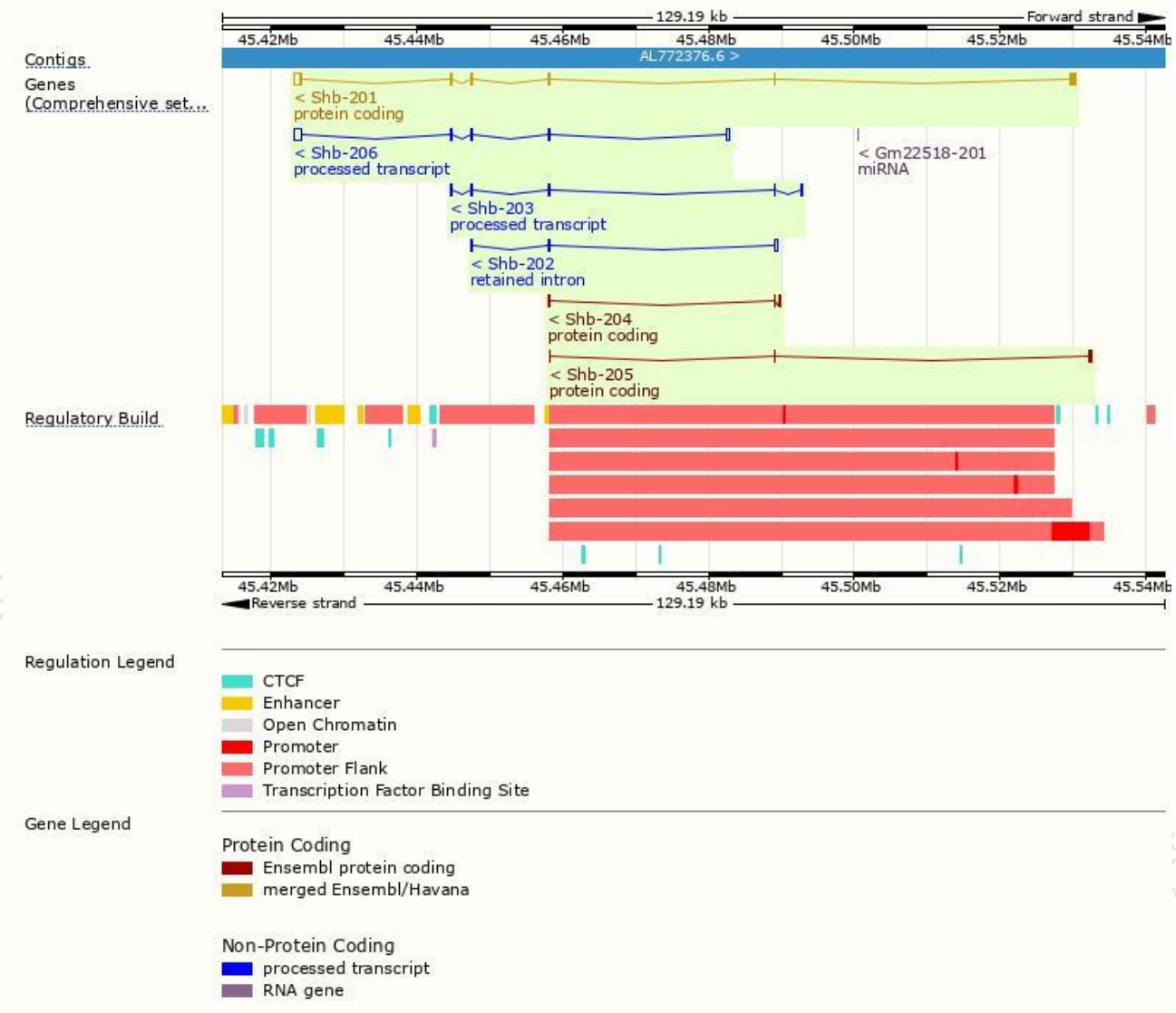
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Shb-201	ENSMUST00000061986.11	2244	503aa	Protein coding	CCDS51173	Q6PD21	TSL:1 GENCODE basic APPRIS P1
Shb-204	ENSMUST00000146236.7	459	145aa	Protein coding	-	A2AKW1	CDS 3' incomplete TSL:3
Shb-205	ENSMUST00000147448.1	368	53aa	Protein coding	-	A2AKW4	CDS 3' incomplete TSL:2
Shb-206	ENSMUST00000147926.7	1609	No protein	Processed transcript	-	-	TSL:1
Shb-203	ENSMUST00000130107.7	726	No protein	Processed transcript	-	-	TSL:3
Shb-202	ENSMUST00000125599.1	737	No protein	Retained intron	-	-	TSL:2

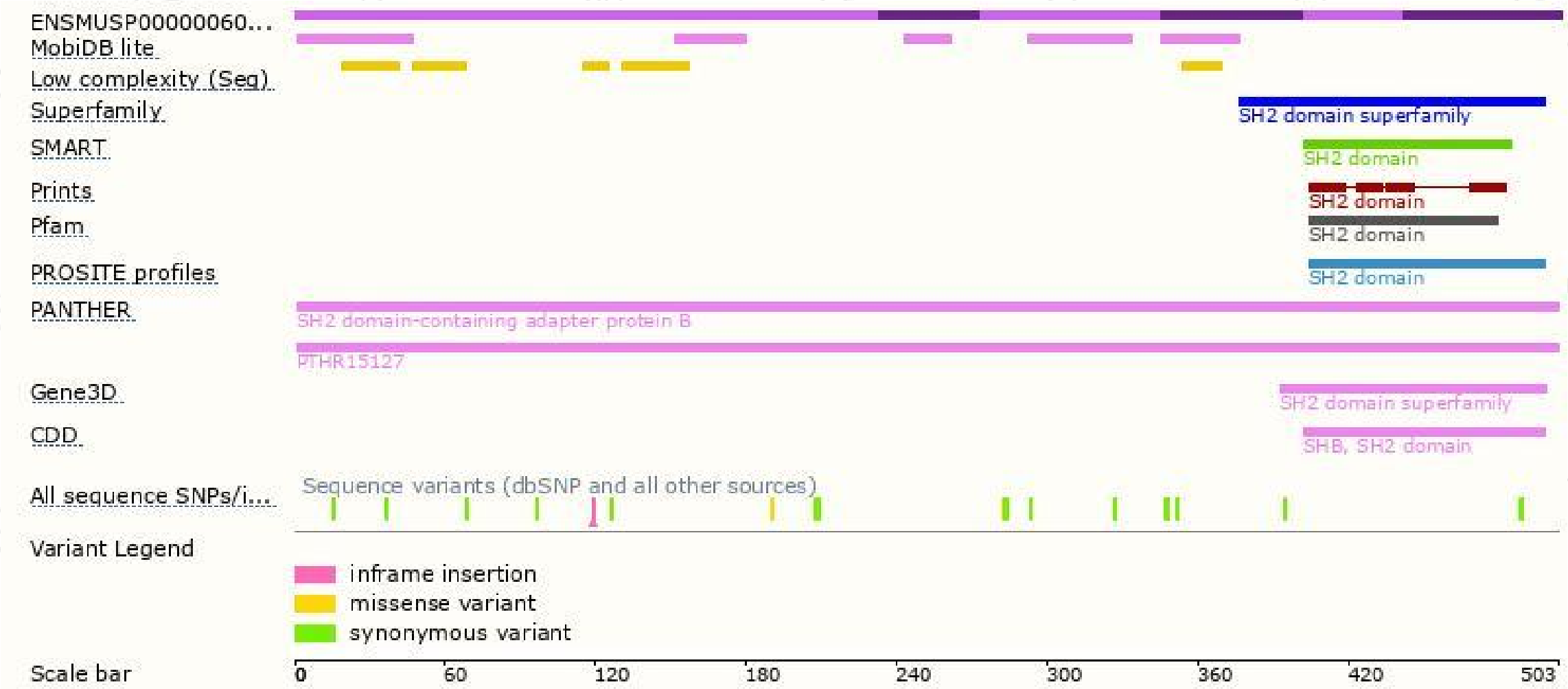
The strategy is based on the design of *Shb-201* transcript,the transcription is shown below:



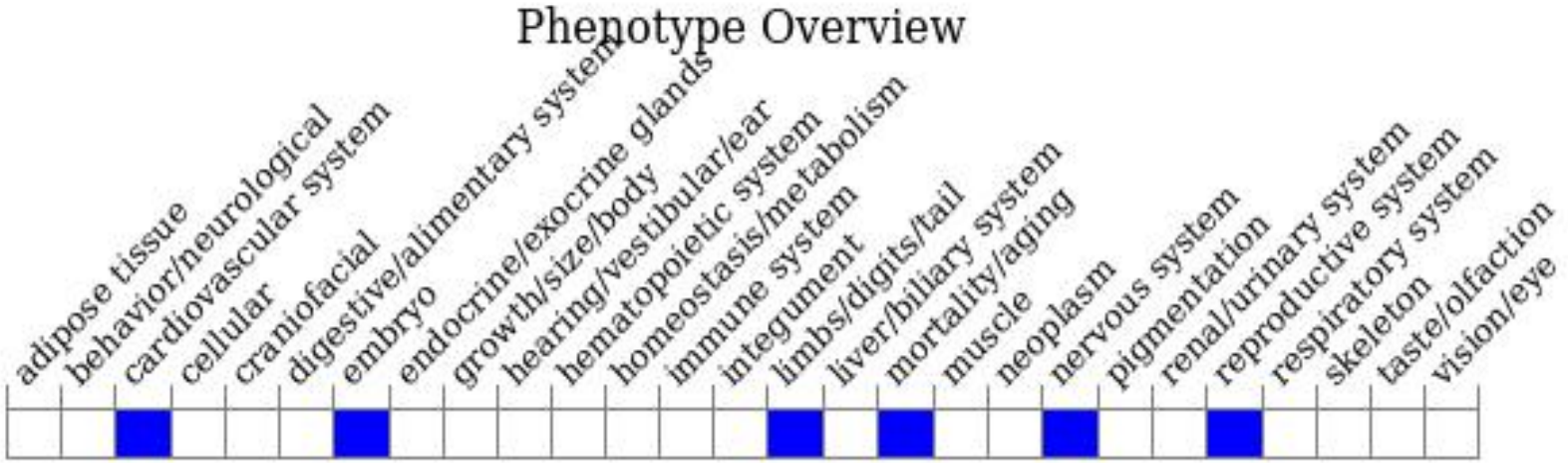
Genomic location distribution



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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