Sh2b1 Cas9-KO Strategy

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Project Overview

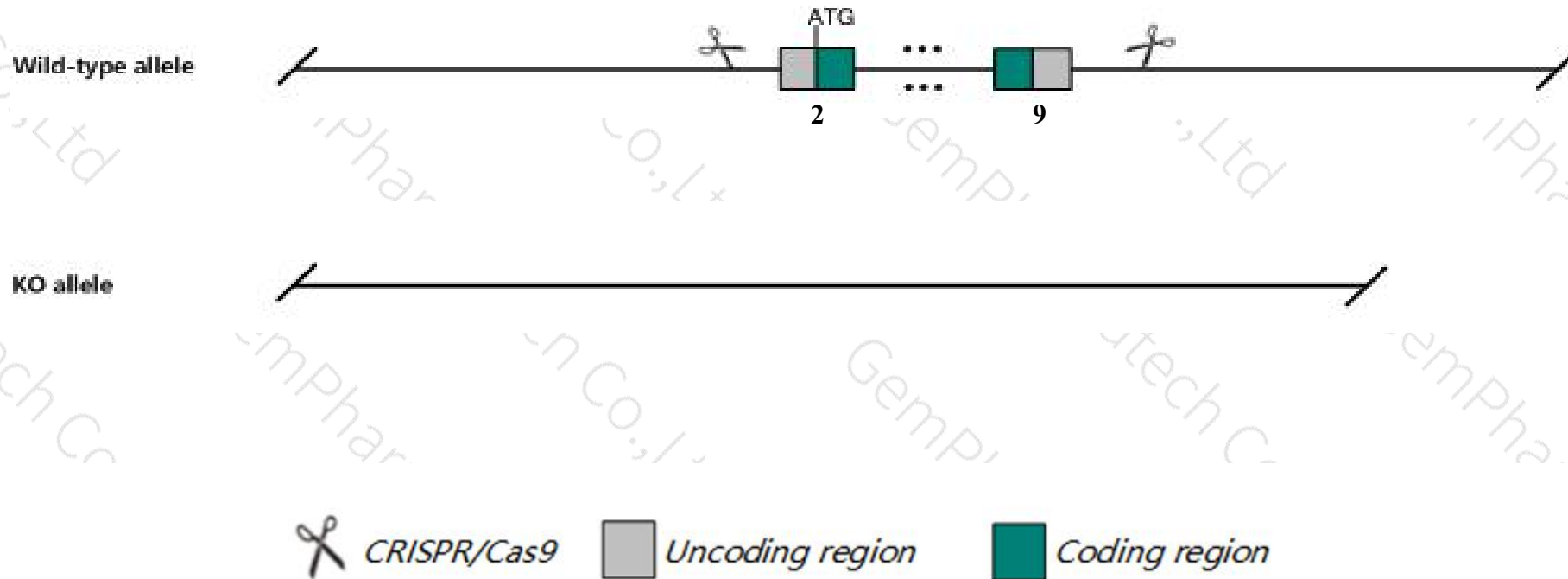|	
Project Name	*Sh2b1*
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sh2b1* gene. The schematic diagram is as follows:

Technical routes

➢ The *Sh2b1* gene has 9 transcripts. According to the structure of *Sh2b1* gene, exon2-exon9 of *Sh2b1-205* (ENSMUST00000205733.1) 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 *Sh2b1* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.

➢ According to the existing MGI data,Homozygous null mice are infertile. Female mice have small, anovulatory ovaries with reduced numbers of follicles and male mice exhibit small testes and sperm deficits. Mice homozygous for a floxed allele activated in the pancreas exhibit impaired glucose homeostasis when fed a high fat diet.

➢ The knockout region is near to the N-terminal of *Atp2a1* gene,this strategy may influence the regulatory function of the N-terminal of *Atp2a1* gene.

➢ The *Sh2b1* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

Sh2b1 SH2B adaptor protein 1 [Mus musculus (house mouse)]

Gene ID: 20399, updated on 31-Jan-2019

Summary

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Official Symbol	Sh2b1 provided by MGI
Official Full Name	SH2B adaptor protein 1 provided by MGI
Primary source	MGI:MGI:1201407
See related	Ensembl:ENSMUSG00000030733
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	AI425885, C530001K22Rik, Irip, Psm, SH2-B, SH2-Bb, Sh2bpsm1, mKIAA1299
Expression	Ubiquitous expression in thymus adult (RPKM 36.7), adrenal adult (RPKM 29.9) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sh2b1-201	ENSMUST00000032978.7	3393	682aa	Protein coding	CCDS40126	Q91ZM2	TSL:1 GENCODE basic APPRIS P3
Sh2b1-202	ENSMUST0000205340.1	3366	670aa	Protein coding	CCDS85412	Q91ZM2	TSL:1 GENCODE basic APPRIS ALT2
Sh2b1-203	ENSMUST0000205440.1	3089	670aa	Protein coding	CCDS85412	Q91ZM2	TSL:1 GENCODE basic APPRIS ALT2
Sh2b1-206	ENSMUST0000205889.1	3011	724aa	Protein coding	CCDS85410	Q91ZM2	TSL:1 GENCODE basic APPRIS ALT2
Sh2b1-205	ENSMUST0000205733.1	2956	756aa	Protein coding	CCDS85411	Q91ZM2	TSL:1 GENCODE basic APPRIS ALT2
Sh2b1-204	ENSMUST0000205497.1	2854	724aa	Protein coding	CCDS85410	Q91ZM2	TSL:1 GENCODE basic APPRIS ALT2
Sh2b1-208	ENSMUST0000206643.1	543	73aa	Protein coding	-	A0A0U1RQ35	CDS 3' incomplete TSL:2
Sh2b1-209	ENSMUST0000206664.1	2909	443aa	Nonsense mediated decay	-	Q91ZM2	TSL:1
Sh2b1-207	ENSMUST0000206515.1	406	No protein	Processed transcript	-	-	TSL:5

The strategy is based on the design of *Sh2b1-205* transcript,The transcription is shown below
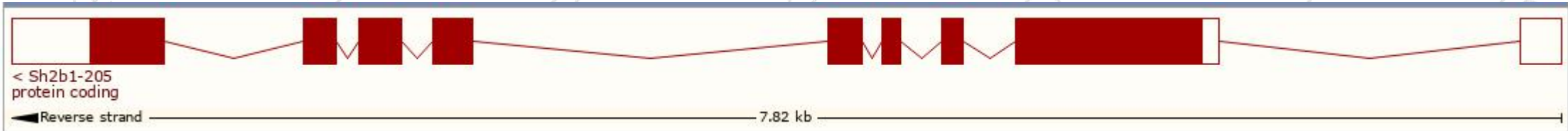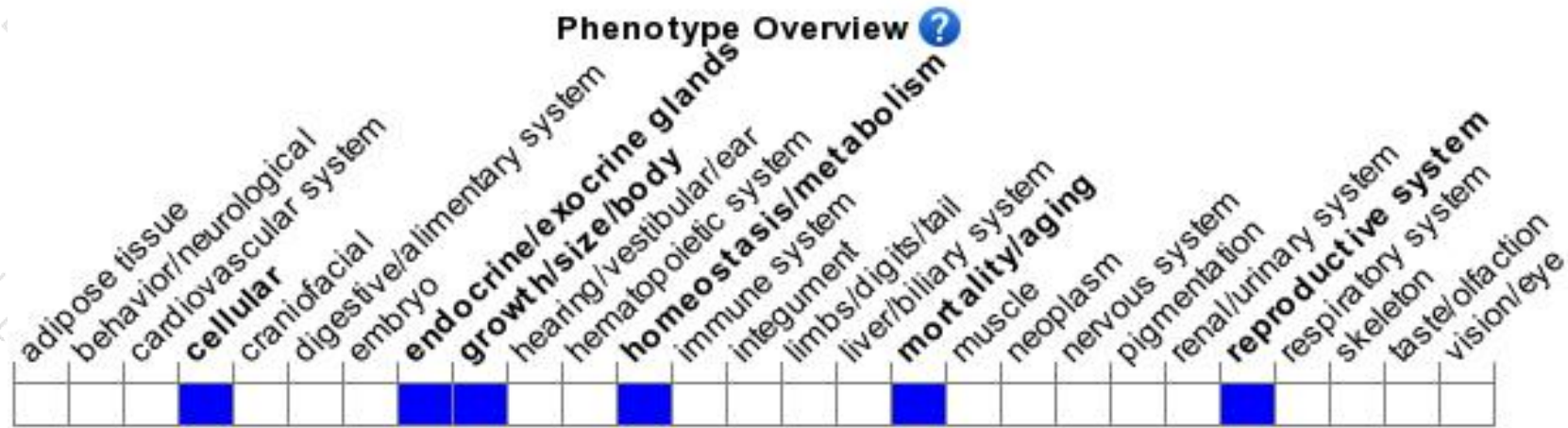
Genomic location distribution
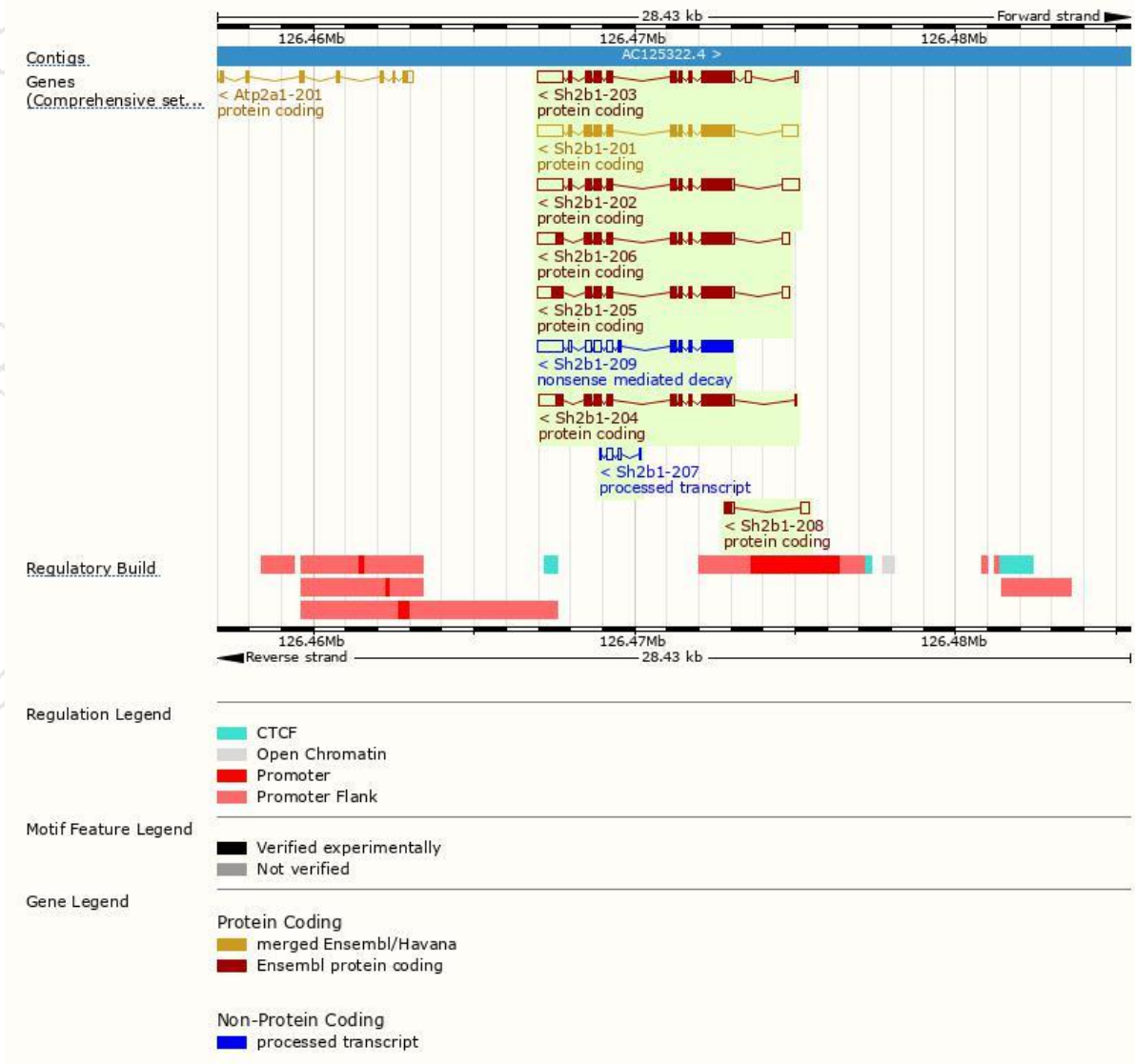
Protein domain

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,Homozygous null mice are infertile. Female mice have small, anovulatory ovaries with reduced numbers of follicles and male mice exhibit small testes and sperm deficits. Mice homozygous for a floxed allele activated in the pancreas exhibit impaired glucose homeostasis when fed a high fat diet.

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