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Sh3rf2 Cas9-CKO Strategy

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Design Date:2024-3-20

Overview

Target Gene Name

- Sh3rf2

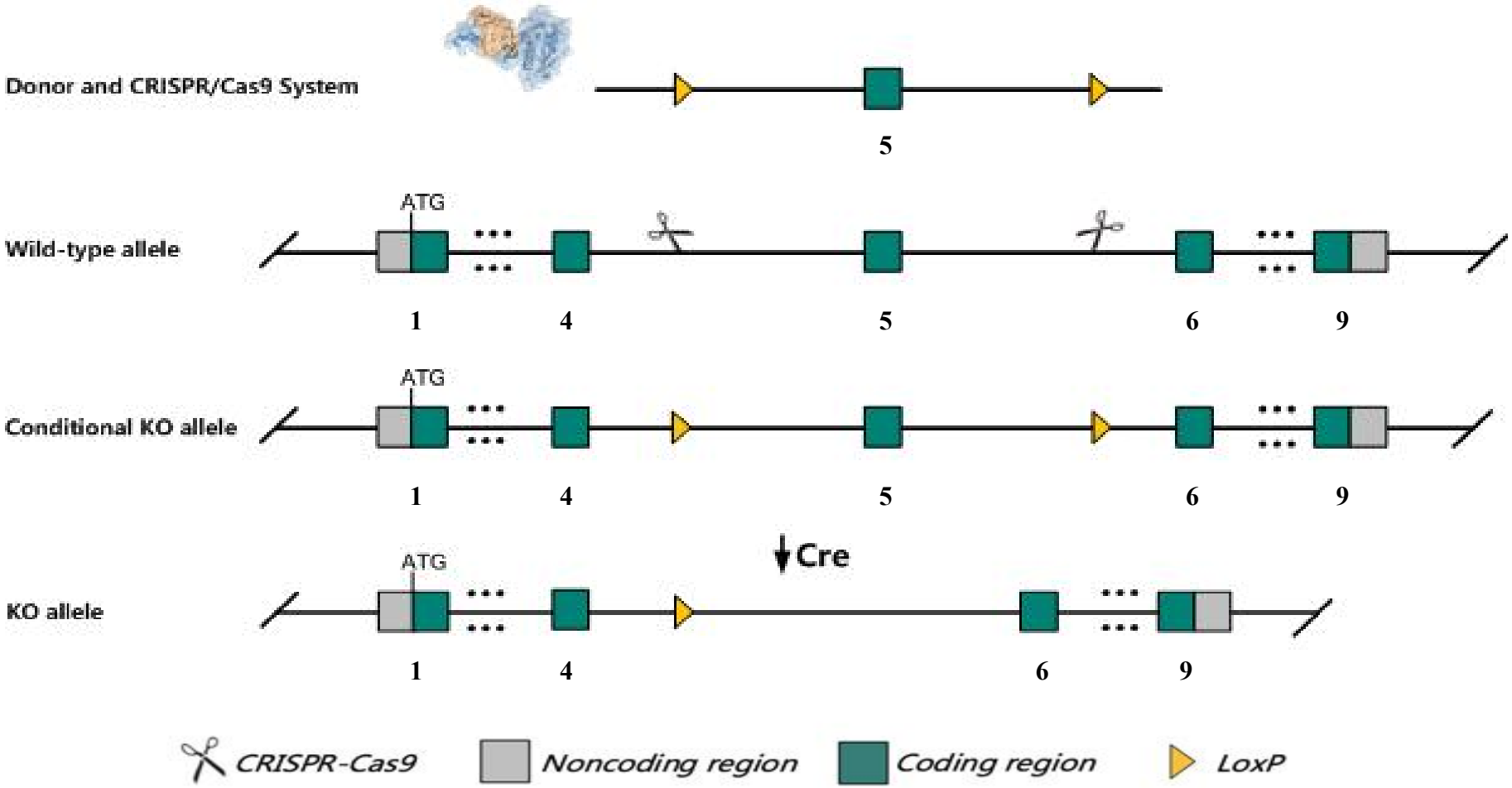
Project Type

- Cas9-CKO

Genetic Background

- C57BL/6JGpt

Strain Strategy

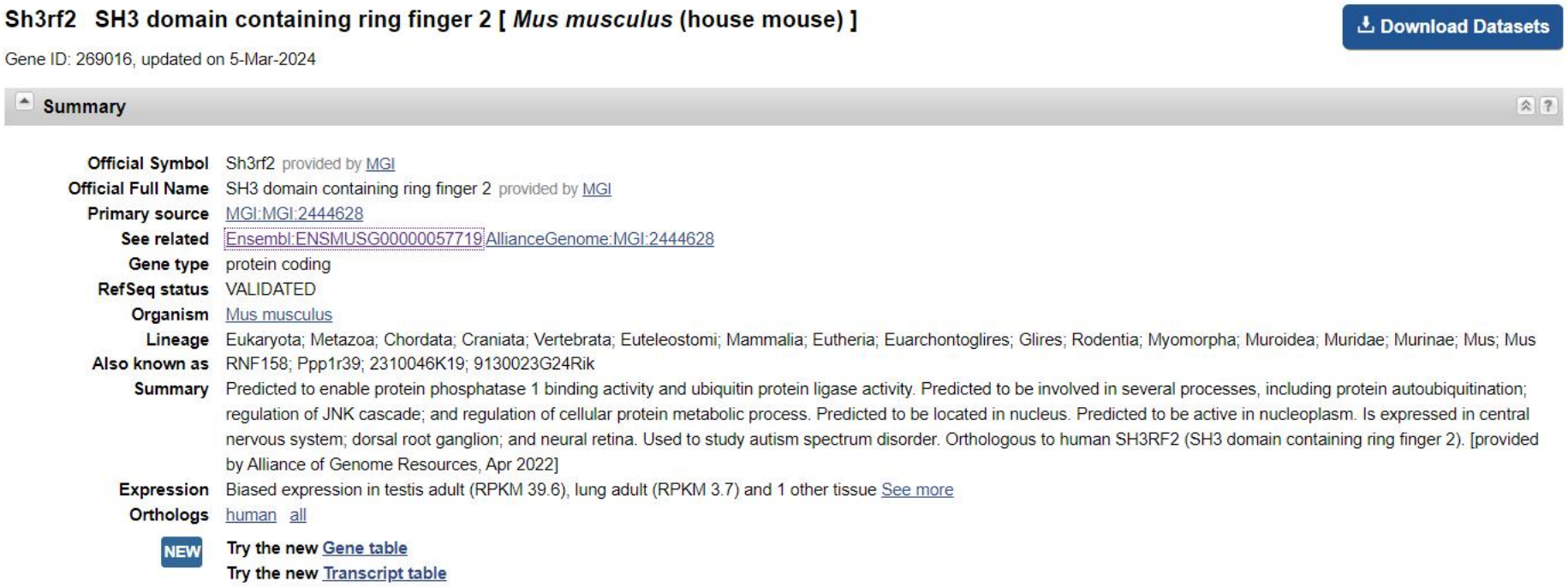


Schematic representation of CRISPR-Cas9 engineering used to edit the *Sh3rf2* gene.

Technical Information

- The *Sh3rf2* gene has 6 transcripts. According to the structure of *Sh3rf2* gene, exon5 of *Sh3rf2*-201 (ENSMUST00000072008.11) transcript is recommended as the knockout region. The region contains 92bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Sh3rf2* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.

Gene Information



Sh3rf2 SH3 domain containing ring finger 2 [*Mus musculus* (house mouse)]

Download Datasets

Gene ID: 269016, updated on 5-Mar-2024

Summary

Official Symbol	Sh3rf2 provided by MGI
Official Full Name	SH3 domain containing ring finger 2 provided by MGI
Primary source	MGI:MGI:2444628
See related	Ensembl:ENSMUSG00000057719 AllianceGenome:MGI:2444628
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	RNF158; Ppp1r39; 2310046K19; 9130023G24Rik
Summary	Predicted to enable protein phosphatase 1 binding activity and ubiquitin protein ligase activity. Predicted to be involved in several processes, including protein autoubiquitination; regulation of JNK cascade; and regulation of cellular protein metabolic process. Predicted to be located in nucleus. Predicted to be active in nucleoplasm. Is expressed in central nervous system; dorsal root ganglion; and neural retina. Used to study autism spectrum disorder. Orthologous to human SH3RF2 (SH3 domain containing ring finger 2). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Biased expression in testis adult (RPKM 39.6), lung adult (RPKM 3.7) and 1 other tissue See more
Orthologs	human all

NEW Try the new Gene table

Try the new Transcript table

Source: https://www.ncbi.nlm.nih.gov/

Transcript Information

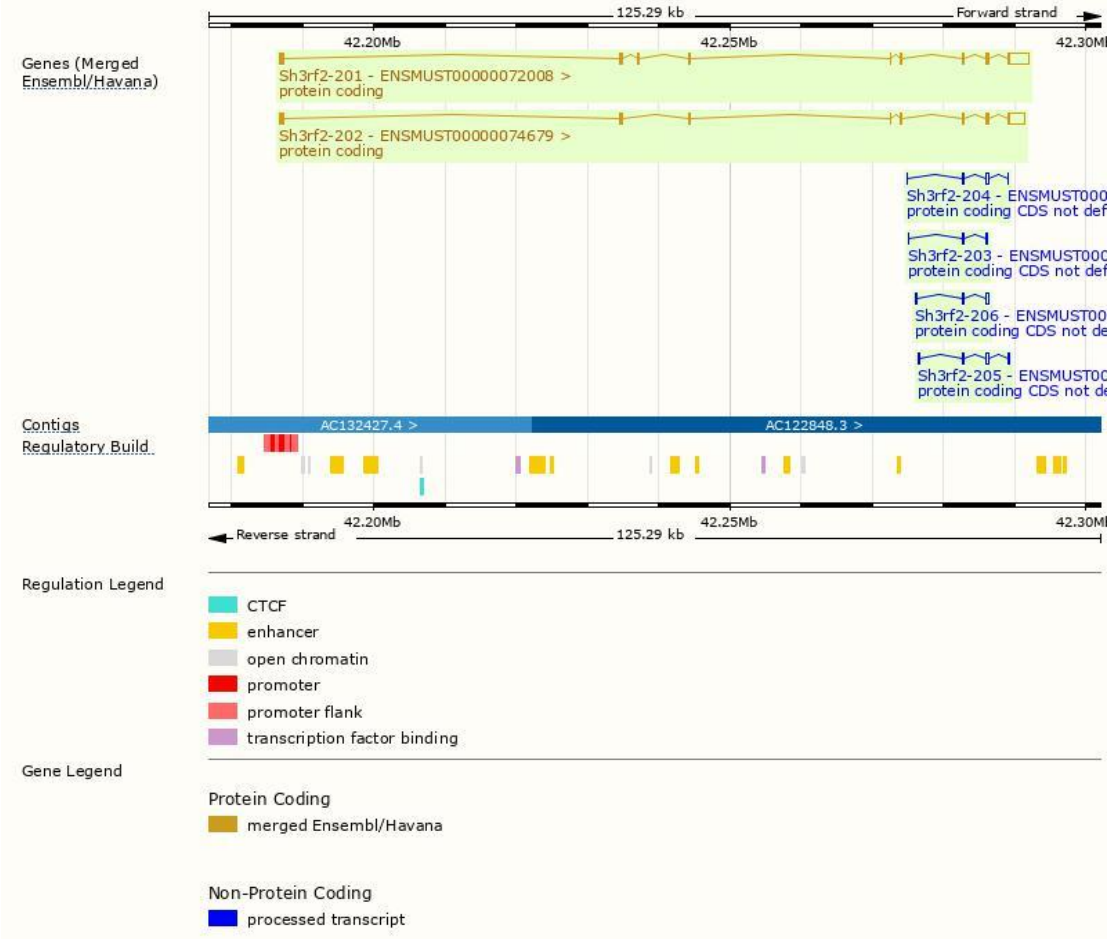
The gene has 6 transcripts,all transcripts are shown below:

Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
ENSMUST00000072008.11	Sh3rf2-201	5092	735aa	Protein coding	CCDS50265	Q8BZT2	Ensembl Canonical GENCODE basic APPRIS P1 TSL:1
ENSMUST00000074679.4	Sh3rf2-202	4444	703aa	Protein coding	CCDS29210	Q8BZT2-2	GENCODE basic TSL:1
ENSMUST00000236947.2	Sh3rf2-205	1143	No protein	Protein coding CDS not defined		-	-
ENSMUST00000132681.8	Sh3rf2-204	836	No protein	Protein coding CDS not defined		-	TSL:2
ENSMUST00000237344.2	Sh3rf2-206	686	No protein	Protein coding CDS not defined		-	-
ENSMUST00000128066.2	Sh3rf2-203	527	No protein	Protein coding CDS not defined		-	TSL:3

The strategy is based on the design of *Sh3rf2*-201 transcript, the transcription is shown below:

Source: https://www.ensembl.org

Genomic Information



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Source: : https://www.ensembl.org

Protein Information

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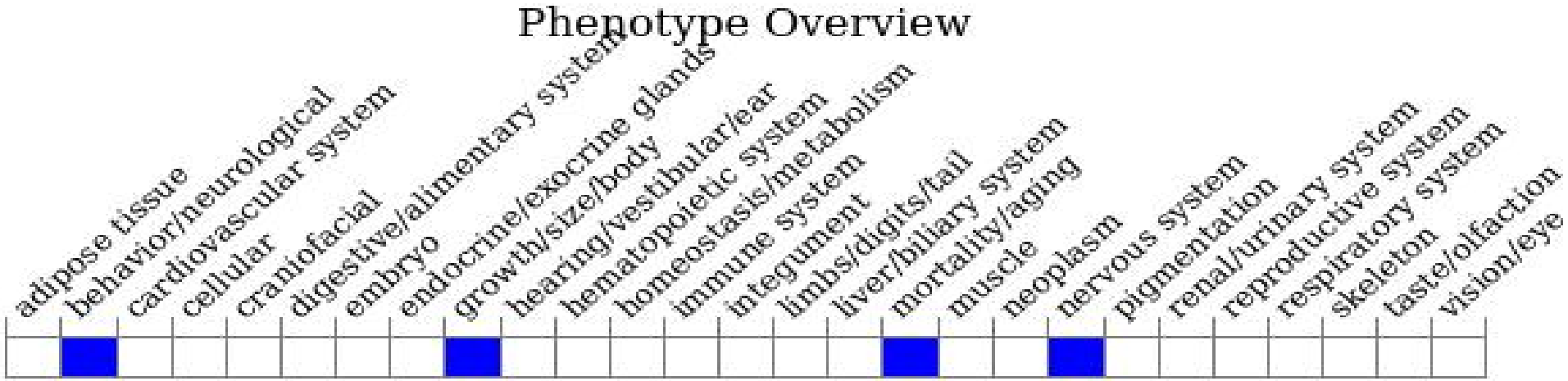
Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)



Source: https://www.informatics.jax.org

Important Information

- *Sh3rf2* is located on Chr18. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.

Reference

Generation of genetically modified mice

Hepatocyte-specific *Sh3rf2* conditional knockout (HKO) mice were generated for this study using the CRISPR/Cas9 system. Briefly, we first designed two single-guide RNAs, namely, sgRNA1 and sgRNA2, that target the upstream and downstream coding sequence (CDS) regions of *Sh3rf2* exon 5 and inserted the sgRNAs into the pUC57-sgRNA vector (Addgene, 51132). Next, we designed an ssODN donor containing two loci of the X-over P1 (loxP) sites and left and right homologous arms. Moreover, the mRNAs of Cas9 and sgRNA were obtained using transcriptional

Yang X, Sun D, Xiang H, Wang S, Huang Y, Li L, Cheng X, Liu H, Hu F, Cheng Y, Ma T, Hu M, Tian H, Tian S, Zhou Y, Zhang P, Zhang XJ, Ji YX, Hu Y, Li H, She ZG. Hepatocyte SH3RF2 Deficiency Is a Key Aggravator for NAFLD. Hepatology. 2021 Sep;74(3):1319-1338. doi: 10.1002/hep.31863. Epub 2021 Aug 30. PMID: 33894019.