

Vps39 Cas9-KO Strategy

Designer: Xueting Zhang

Reviewer: Ruirui Zhang

Design Date: 2023-10-31

Overview

Target Gene Name

- Vps39

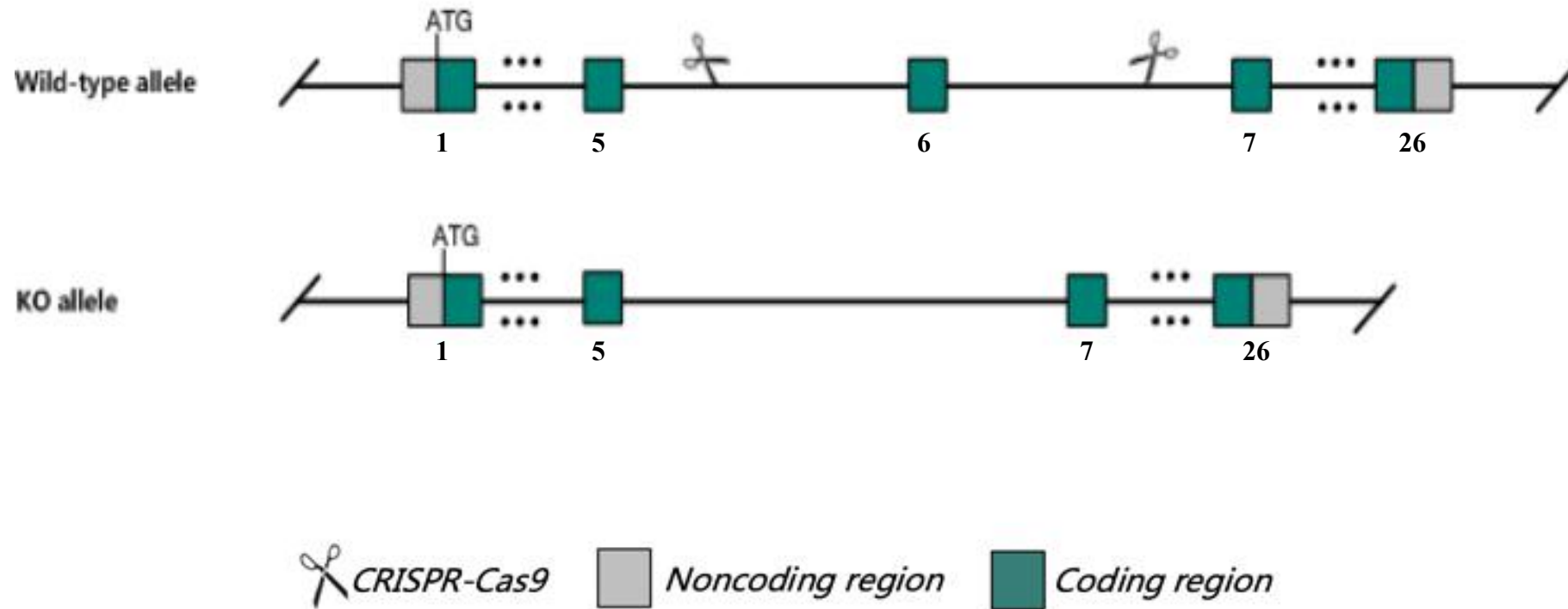
Project Type

- Cas9-KO

Genetic Background

- C57BL/6JGpt

Strain Strategy



Technical Information

- The *Vps39* gene has 10 transcripts. According to the structure of *Vps39* gene, exon6 of *Vps39*-202 (ENSMUST00000102501.10) transcript is recommended as the knockout region. The region contains 95bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Vps39* gene. The brief process is as follows: gRNAs were transcribed in vitro. Cas9 and gRNAs 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.

Gene Information

Vps39 VPS39 HOPS complex subunit [Mus musculus (house mouse)]

Gene ID: 269338, updated on 31-May-2023

Summary

Official Symbol	Vps39 provided by MGI
Official Full Name	VPS39 HOPS complex subunit provided by MGI
Primary source	MGI:MGI:2443189
See related	Ensembl:ENSMUSG00000027291
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	A230065P22Rik, Vam6, Vam6P, mVam6
Summary	Predicted to be involved in endocytic recycling; lysosomal transport; and vesicle fusion. Predicted to act upstream of or within protein transport. Part of AP-3 adaptor complex. Is expressed in several structures, including brain; genitourinary system; gut; quadriceps femoris muscle; and trigeminal nerve. Orthologous to human VPS39 (VPS39 subunit of HOPS complex). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Ubiquitous expression in CNS E18 (RPKM 15.5), whole brain E14.5 (RPKM 15.5) and 28 other tissues See more
Orthologs	human all

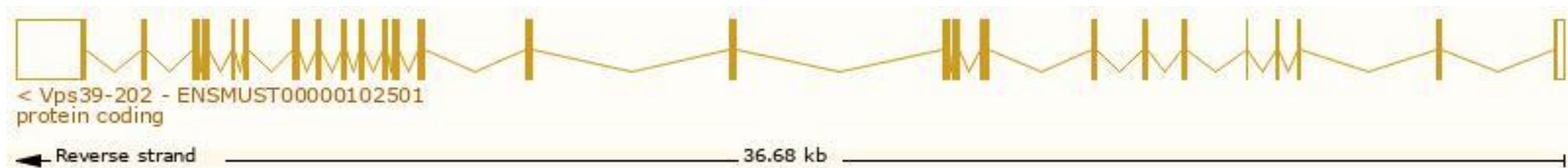
Source: <https://www.ncbi.nlm.nih.gov/>

Transcript Information

The gene has 10 transcripts, all transcripts are shown below:

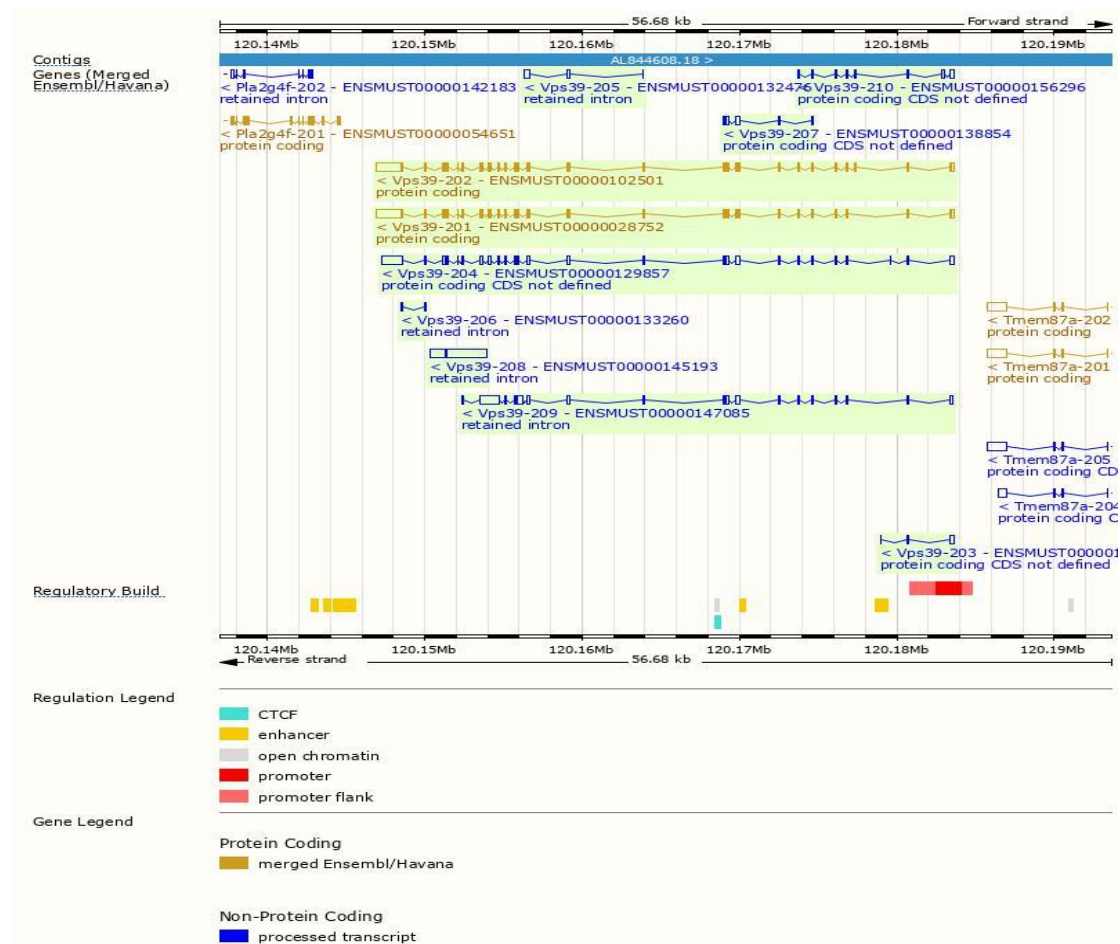
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
ENSMUST00000102501.10	Vps39-202	4384	886aa	Protein coding	CCDS38210	Q8R5L3-1	Ensembl Canonical Gencode basic APPRIS ALT1 TSL:1
ENSMUST00000028752.8	Vps39-201	4339	875aa	Protein coding	CCDS16619	Q8R5L3-2	Gencode basic APPRIS P4 TSL:1
ENSMUST00000129857.8	Vps39-204	4055	No protein	Protein coding CDS not defined		-	TSL:1
ENSMUST00000156296.8	Vps39-210	691	No protein	Protein coding CDS not defined		-	TSL:3
ENSMUST00000138854.2	Vps39-207	602	No protein	Protein coding CDS not defined		-	TSL:5
ENSMUST00000126526.2	Vps39-203	343	No protein	Protein coding CDS not defined		-	TSL:3
ENSMUST00000145193.2	Vps39-208	3545	No protein	Retained intron		-	TSL:1
ENSMUST00000147085.8	Vps39-209	3379	No protein	Retained intron		-	TSL:5
ENSMUST00000132476.2	Vps39-205	469	No protein	Retained intron		-	TSL:3
ENSMUST00000133260.2	Vps39-206	200	No protein	Retained intron		-	TSL:3

The strategy is based on the design of *Vps39-202* transcript, the transcription is shown below:

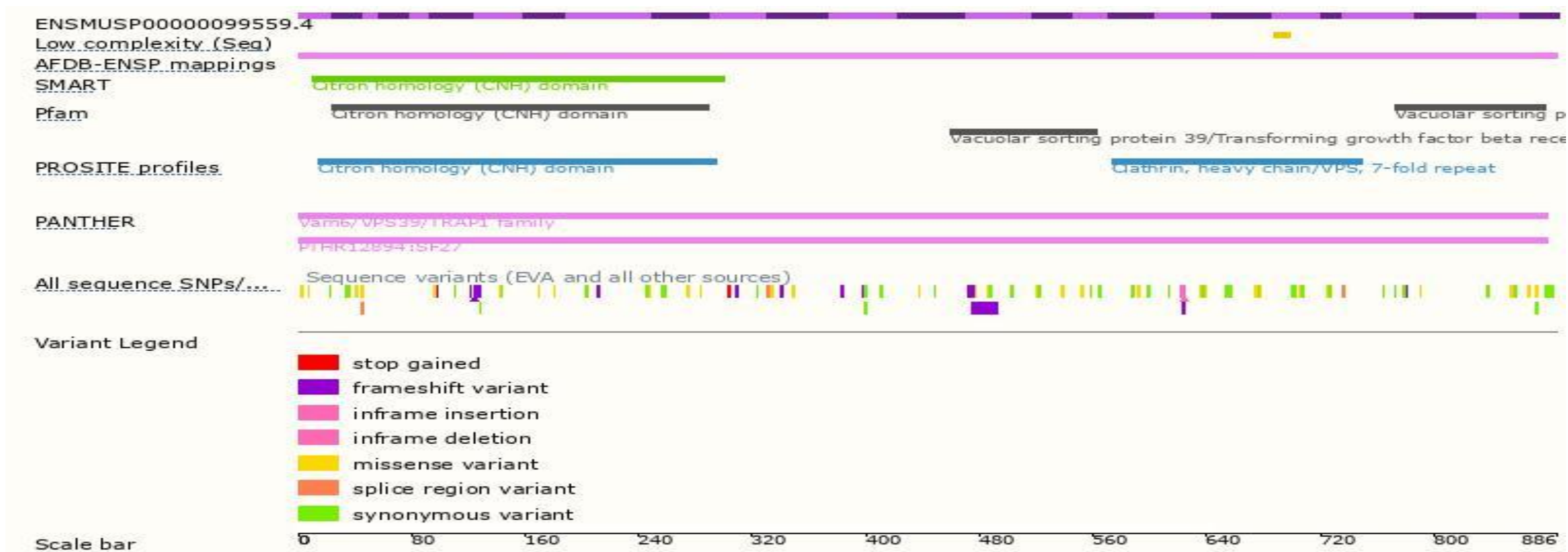


Source: <https://www.ensembl.org>

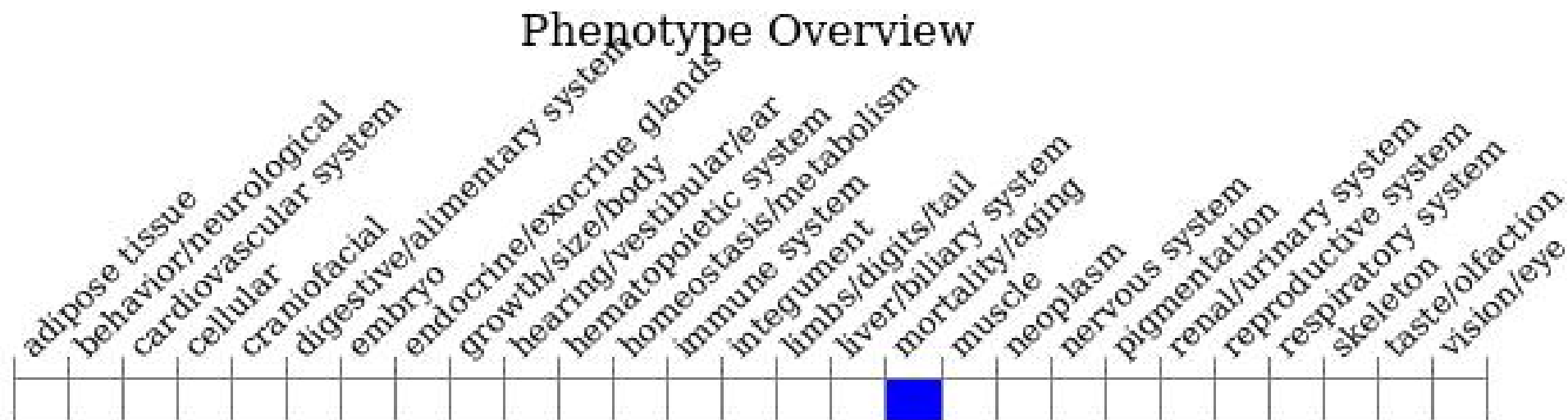
Genomic Information



Protein Information



Mouse Phenotype Information (MGI)



- Mice exhibit embryonic lethality between E2.5 and E7.5.

Important Information

- According to the existing MGI data, mice exhibit embryonic lethality between E2.5 and E7.5.
- The some amino acids of N-terminal of *Vps39* will be remained, and the effect is unknown.
- *Vps39* is located on Chr2. 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 risks of the mutation on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Reference

<https://www.mousephenotype.org/data/genes/MGI:2443189>

Vps39^{tm1a(EUCOMM)Hmgu} KO first allele (reporter-tagged insertion with conditional potential) Mice, Targeting vectors, ES Cells

1 2 3 5 FRT lacZ neo FRT loxP 6 loxP 7 9 10 12

SA pA

Mice (1) ES Cells (19) Targeting Vectors (2) Tissues (0)

Colony Name	Genetic Background	Production Centre	QC Data	ES Cell/Parent Mouse Colony}	Order / Contact
HEPD0537_1_E10	C57BL/6NTac	RIKEN BRC	QC data	HEPD0537_1_E10	Contact RIKEN BRC