

Vti1a Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Yanhua Shen

Date:2020-02-14

Project Overview



Project Name

Vti1a

Project type

Cas9-CKO

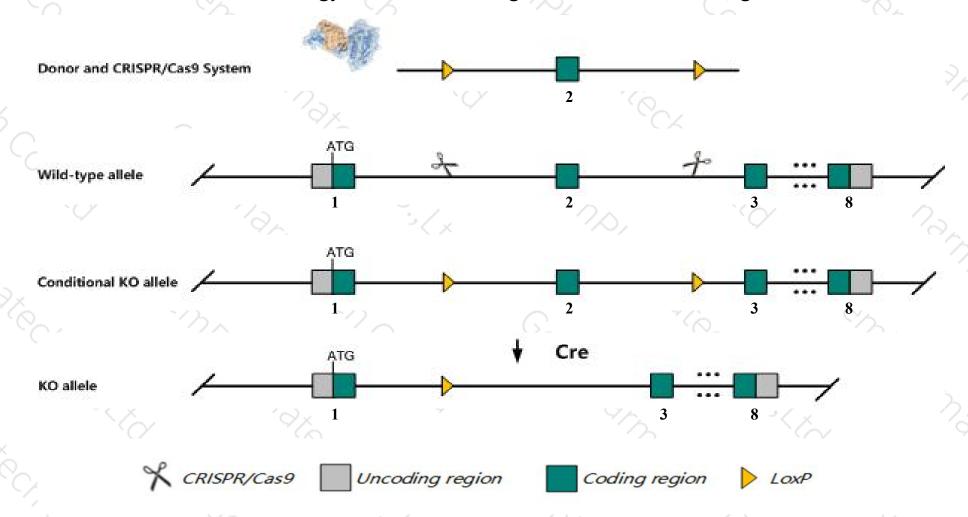
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Vti1a* gene. The schematic diagram is as follows:



Technical routes



- The *Vti1a* gene has 7 transcripts. According to the structure of *Vti1a* gene, exon2 of *Vti1a-201*(ENSMUST00000095950.2) transcript is recommended as the knockout region. The region contains 59bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Vti1a* 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



- > According to the existing MGI data, Mice homozygous for a knock-out allele are viable and fertile.
- ➤ Transcript *Vti1a*-203 may not be affected.
- The floxed region is near to the N-terminal of *Zdhhc6* gene, this strategy may influence the regulatory function of the N-terminal of *Zdhhc6* gene.
- The *Vti1a* gene is located on the Chr19. 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)



Vti1a vesicle transport through interaction with t-SNAREs 1A [Mus musculus (house mouse)]

Gene ID: 53611, updated on 26-Nov-2019

Summary

☆ ?

Official Symbol Vti1a provided by MGI

Official Full Name vesicle transport through interaction with t-SNAREs 1A provided by MGI

Primary source MGI:MGI:1855699

See related Ensembl:ENSMUSG00000024983

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as Vti1; MVti1; MVti1a; Vti1-rp2; 1110014F16Rik; 1110018K19Rik; 4921537J05Rik

Expression Ubiquitous expression in cortex adult (RPKM 1.7), frontal lobe adult (RPKM 1.7) and 28 other tissues See more

Orthologs human all

Genomic context



Location: 19; 19 D2

See Vti1a in Genome Data Viewer

Exon count: 16

| Annotation release | Status | Assembly | Chr | Location |
|--------------------|-------------------|------------------------------|-----|--------------------------------|
| 108 | current | GRCm38.p6 (GCF_000001635.26) | 19 | NC_000085.6 (5531603055627461) |
| Build 37.2 | previous assembly | MGSCv37 (GCF_000001635.18) | 19 | NC_000085.5 (5539084155701051) |

Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

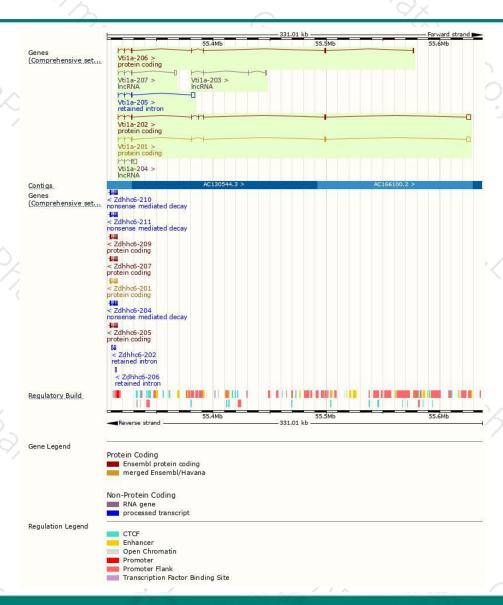
| Name | Transcript ID | bp | Protein | Biotype | ccds | UniProt | Flags |
|-----------|----------------------|------|--------------|-----------------|-----------|----------------|-------------------------------|
| Vti1a-201 | ENSMUST00000095950.2 | 3405 | 217aa | Protein coding | CCDS29910 | <u>089116</u> | TSL:1 GENCODE basic APPRIS P2 |
| Vti1a-202 | ENSMUST00000223690.1 | 4175 | 224aa | Protein coding | - | Q5FWJ7 | GENCODE basic APPRIS ALT1 |
| Vti1a-206 | ENSMUST00000225529.1 | 1244 | <u>197aa</u> | Protein coding | - | A0A286YDF2 | GENCODE basic |
| Vti1a-205 | ENSMUST00000225392.1 | 3370 | No protein | Retained intron | 22 | 20 | |
| Vti1a-204 | ENSMUST00000225051.1 | 2919 | No protein | IncRNA | - | 7.0 | |
| Vti1a-207 | ENSMUST00000226069.1 | 1599 | No protein | IncRNA | - | - 8 | |
| Vti1a-203 | ENSMUST00000224396.1 | 955 | No protein | IncRNA | | -8 | |

The strategy is based on the design of Vti1a-201 transcript, The transcription is shown below



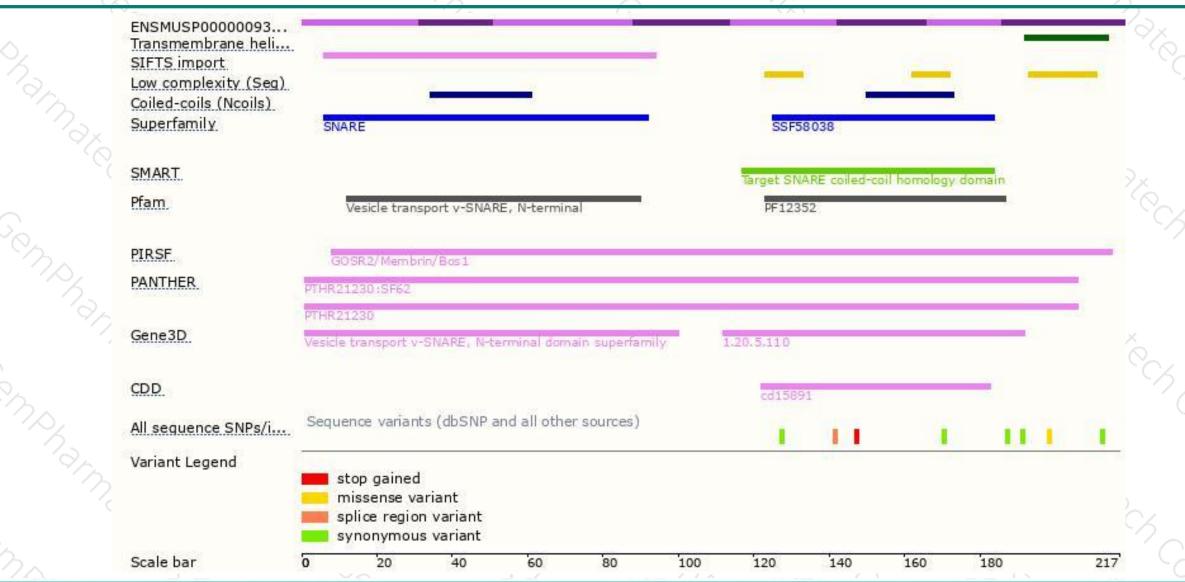
Genomic location distribution





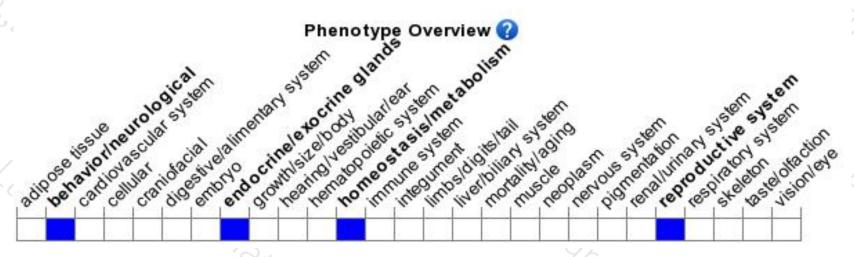
Protein domain





Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Mice homozygous for a knock-out allele are viable and fertile.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





