

# Slc6a9 Cas9-KO Strategy

**Designer: Rui Xiong** 

**Reviewer: Yumeng Wang** 

**Design Date: 2022-1-7** 

## **Project Overview**

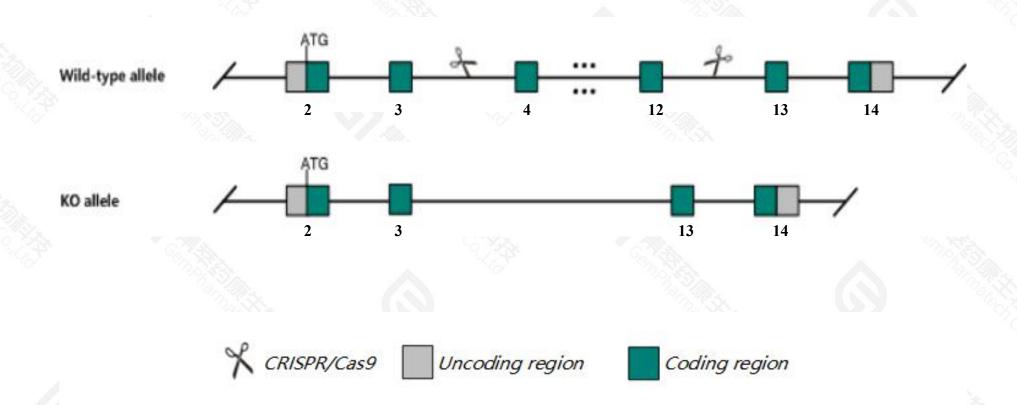


Project Name	Slc6a9
Project type	Cas9-KO
Strain background	C57BL/6JGpt

## **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the Slc6a9 gene. The schematic diagram is as follows:



#### **Technical routes**



- The *Slc6a9* gene has 12 transcripts. According to the structure of *Slc6a9* gene, exon4-exon12 of *Slc6a9*201(ENSMUST0000030269.14) transcript is recommended as the knockout region. The region contains 1349bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Slc6a9* 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**



- $\triangleright$  The partial intron of Gm17114 gene will be deleted together after Cre recombination in this strategy.
- > Transcript Slc6a9-208 may not be affected.
- > According to the existing MGI data, homozygous null mice die shortly after birth exhibiting breathing and movement deficiencies.
- > The *Slc6a9* 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)



## SIc6a9 solute carrier family 6 (neurotransmitter transporter, glycine), member 9 [Mus musculus (house mouse)]

Gene ID: 14664, updated on 2-Mar-2021

#### Summary



Official Symbol Slc6a9 provided by MGI

Official Full Name solute carrier family 6 (neurotransmitter transporter, glycine), member 9 provided by MGI

Primary source MGI:MGI:95760

See related Ensembl:ENSMUSG00000028542

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 Glyt, Glyt-, Glyt-1, Glyt1

Expression Ubiquitous expression in liver E14.5 (RPKM 33.4), cerebellum adult (RPKM 30.9) and 27 other tissuesSee more

Orthologs <u>human</u> all

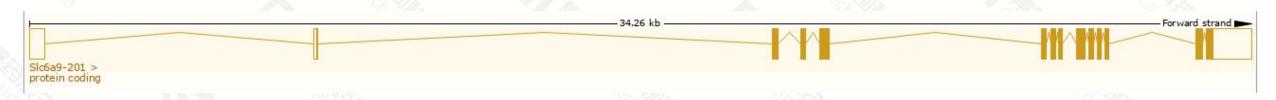
## Transcript information (Ensembl)



The gene has 12 transcripts, all transcripts are shown below:

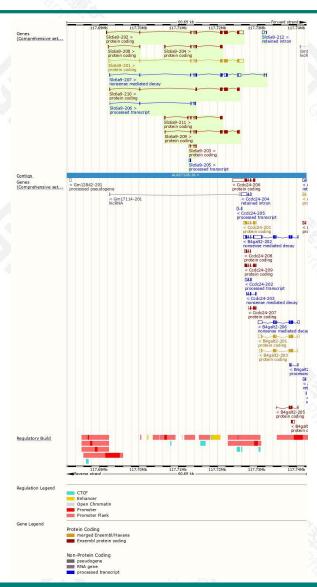
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Slc6a9-201	ENSMUST00000030269.14	3479	<u>633aa</u>	Protein coding	CCDS18538		TSL:1 , GENCODE basic , APPRIS P1
Slc6a9-202	ENSMUST00000063857.11	3146	<u>633aa</u>	Protein coding	CCDS18538		TSL:1 , GENCODE basic , APPRIS P1
Slc6a9-209	ENSMUST00000163288.2	2107	<u>637aa</u>	Protein coding	12		TSL:5 , GENCODE basic ,
Slc6a9-210	ENSMUST00000169885.8	1876	521aa	Protein coding			TSL:5 , GENCODE basic ,
Slc6a9-211	ENSMUST00000169990.8	1344	<u>373aa</u>	Protein coding	<u>a</u>		CDS 3' incomplete , TSL:5 ,
Slc6a9-204	ENSMUST00000132043.9	1194	321aa	Protein coding	-		CDS 3' incomplete , TSL:5 ,
Slc6a9-203	ENSMUST00000123994.3	456	116aa	Protein coding	3 <del>.</del>		CDS 3' incomplete , TSL:5 ,
Slc6a9-208	ENSMUST00000151316.2	244	<u>laa</u>	Protein coding	12		CDS 3' incomplete , TSL:5 ,
Slc6a9-207	ENSMUST00000149168.2	2834	126aa	Nonsense mediated decay	8		TSL:1,
Slc6a9-206	ENSMUST00000136476.2	488	No protein	Processed transcript	34		TSL:5,
Slc6a9-205	ENSMUST00000135769.2	370	No protein	Processed transcript	12		TSL:2,
Slc6a9-212	ENSMUST00000170733.2	678	No protein	Retained intron			TSL:3,
	UV	-					TVL WHILE

The strategy is based on the design of *Slc6a9-201* transcript, the transcription is shown below:



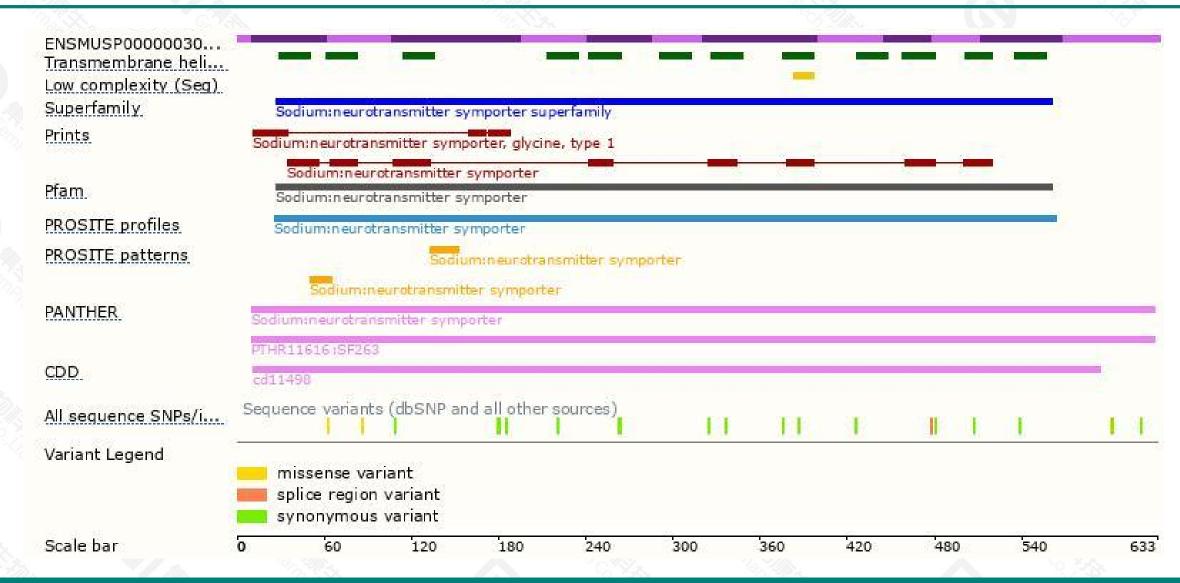
#### Genomic location distribution





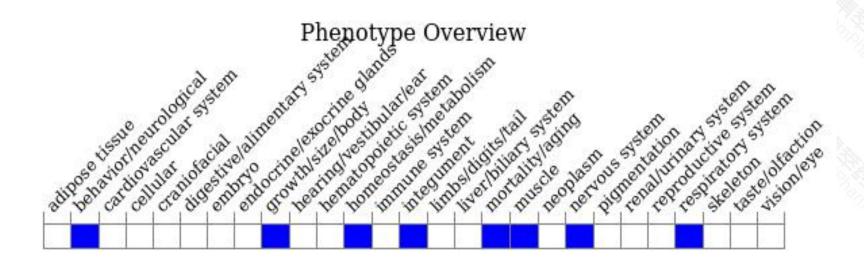
#### Protein domain





## 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, homozygous null mice die shortly after birth exhibiting breathing and movement deficiencies.



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

Tel: 400-9660890





