

Slc32a1 Cas9-CKO Strategy

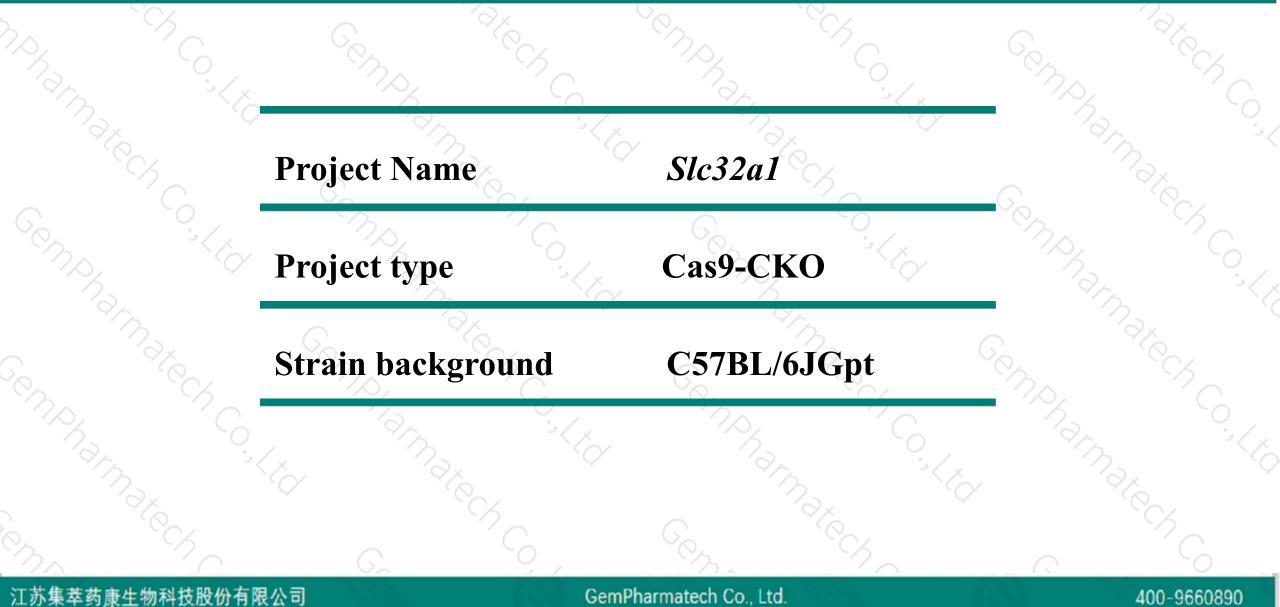
Designer: Reviewer:

Design Date:

Huan Wang Huan Fan 2019-12-17

Project Overview



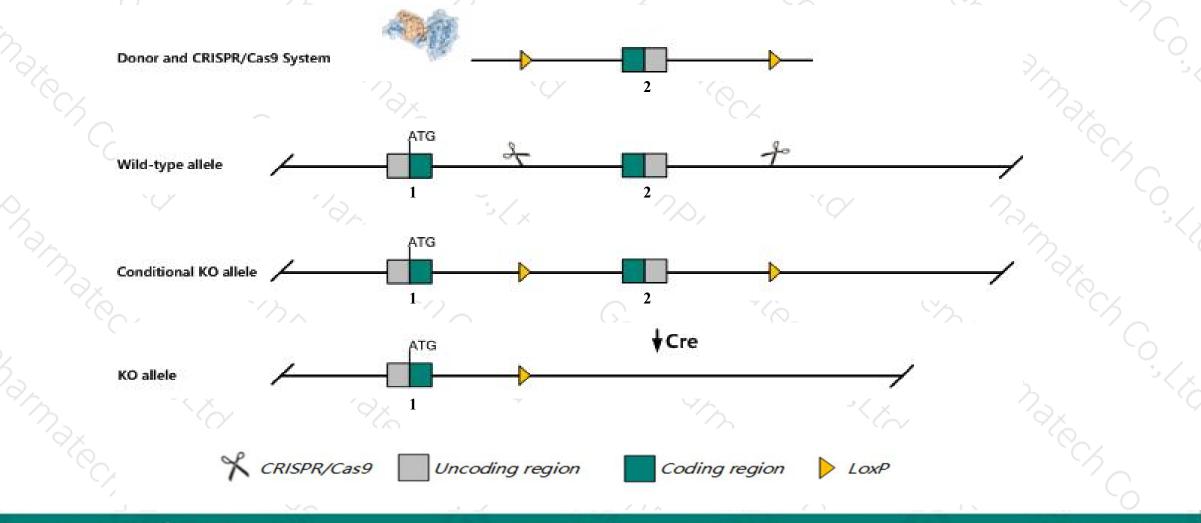


Conditional Knockout strategy



400-9660890

This model will use CRISPR/Cas9 technology to edit the *Slc32a1* gene. The schematic diagram is as follows:



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The Slc32a1 gene has 1 transcript. According to the structure of Slc32a1 gene, exon2 of Slc32a1-201 (ENSMUST00000045738.4) transcript is recommended as the knockout region. The region contains most 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 *Slc32a1* 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.



- According to the existing MGI data, Homozygous null mice been independently reported to die perinatally exhibiting a hunched posture, respiratory failure, cleft secondary palate due to failure of palate shelf elevation, umbilical hernia or omphalocele, and loss of neurotransmitter release in both GABAergic and glycinergic neurons.
- The Slc32a1 gene is located on the Chr2. 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.

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Gene information (NCBI)



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SIc32a1 solute carrier family 32 (GABA vesicular transporter), member 1 [Mus musculus (house mouse)]

Gene ID: 22348, updated on 9-Apr-2019

Summary

Official Symbol	SIc32a1 provided by MGI
Official Full Name	solute carrier family 32 (GABA vesicular transporter), member 1 provided by MGI
Primary source	MGI:MGI:1194488
See related	Ensembl:ENSMUSG0000037771
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	R75019, VGAT, Viaat
Expression	Biased expression in cerebellum adult (RPKM 56.5), frontal lobe adult (RPKM 42.0) and 5 other tissues See more
Orthologs	human all

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The gene has 1 transcript, and the transcript is shown below:

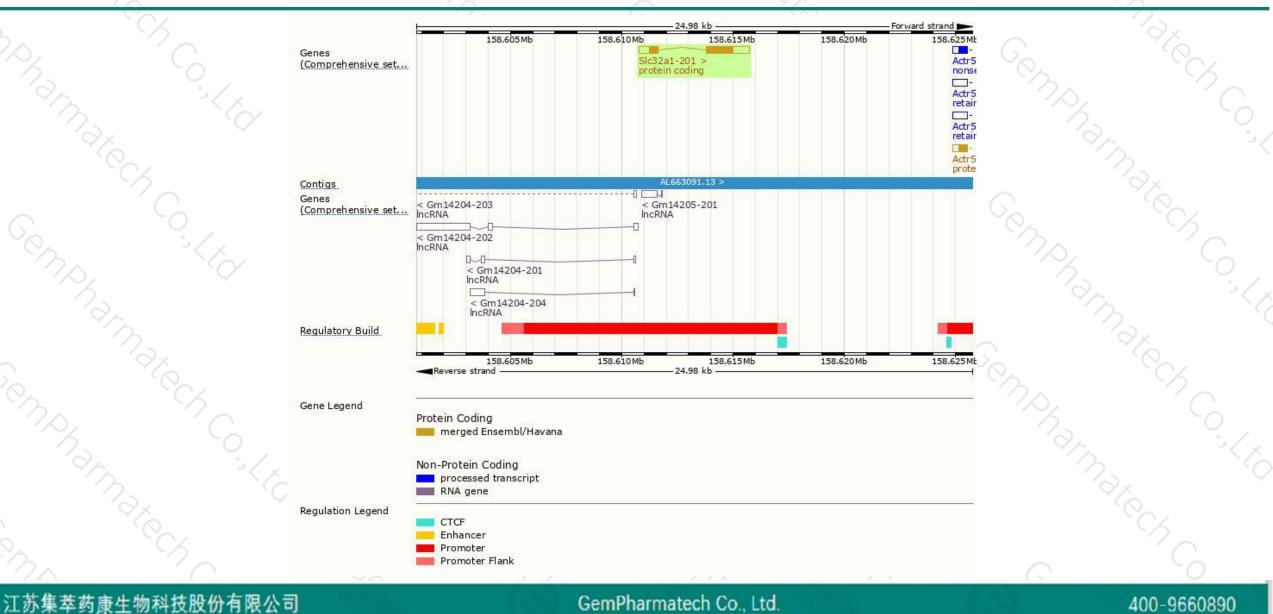
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
SIc32a1-201	ENSMUST00000045738.4	2797	<u>525aa</u>	Protein coding	CCDS38309	<u>035633 Q49598</u>	TSL:1 GENCODE basic APPRIS P1
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The strategy	is based on the design of	f <i>Slc3.</i>	2a1-201	transcript,The	transcription	n is shown below	Conpt Co
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Slc32a1-201 > protein coding							
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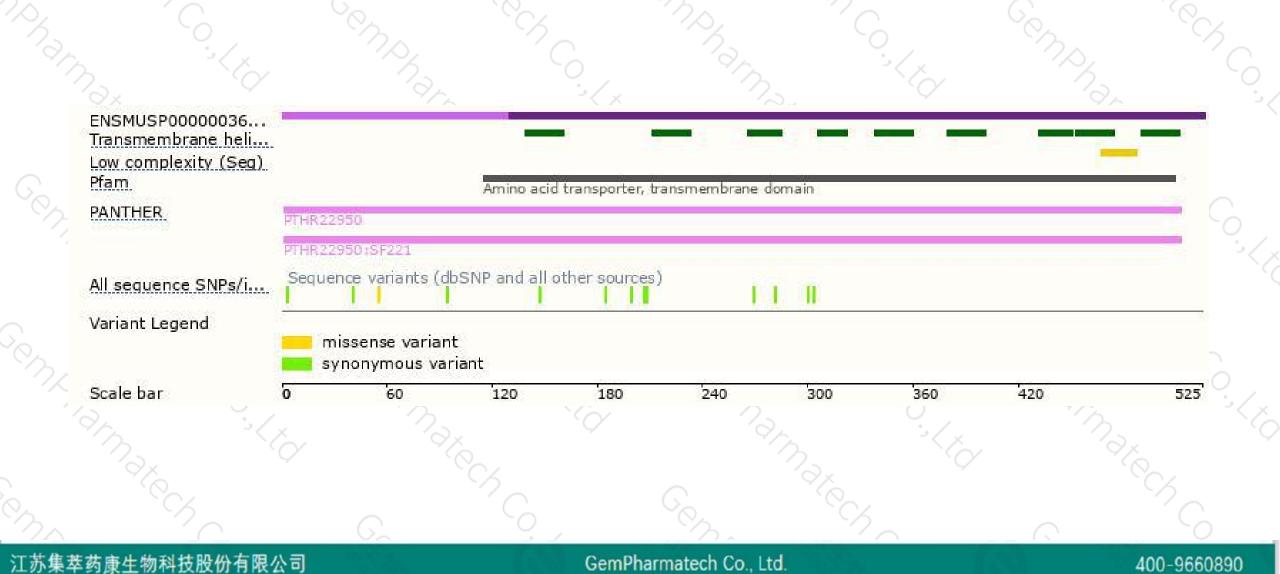
Genomic location distribution





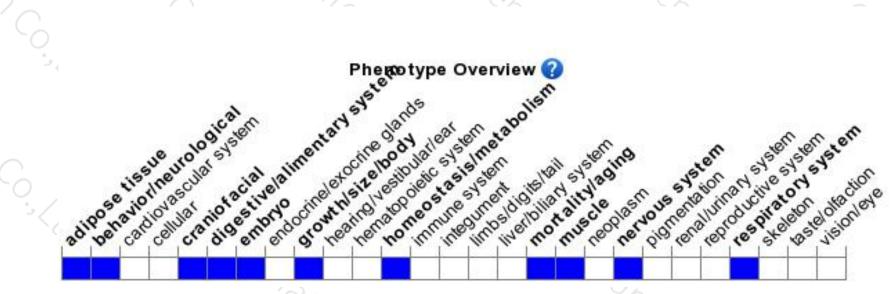
Protein domain





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 been independently reported to die perinatally exhibiting a hunched posture, respiratory failure, cleft secondary palate due to failure of palate shelf elevation, umbilical hernia or omphalocele, and loss of neurotransmitter release in both GABAergic and glycinergic neurons.

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



