Syt2 Cas9-KO Strategy

Designer: Jiayuan Yao

Reviewer: Longyun Hu

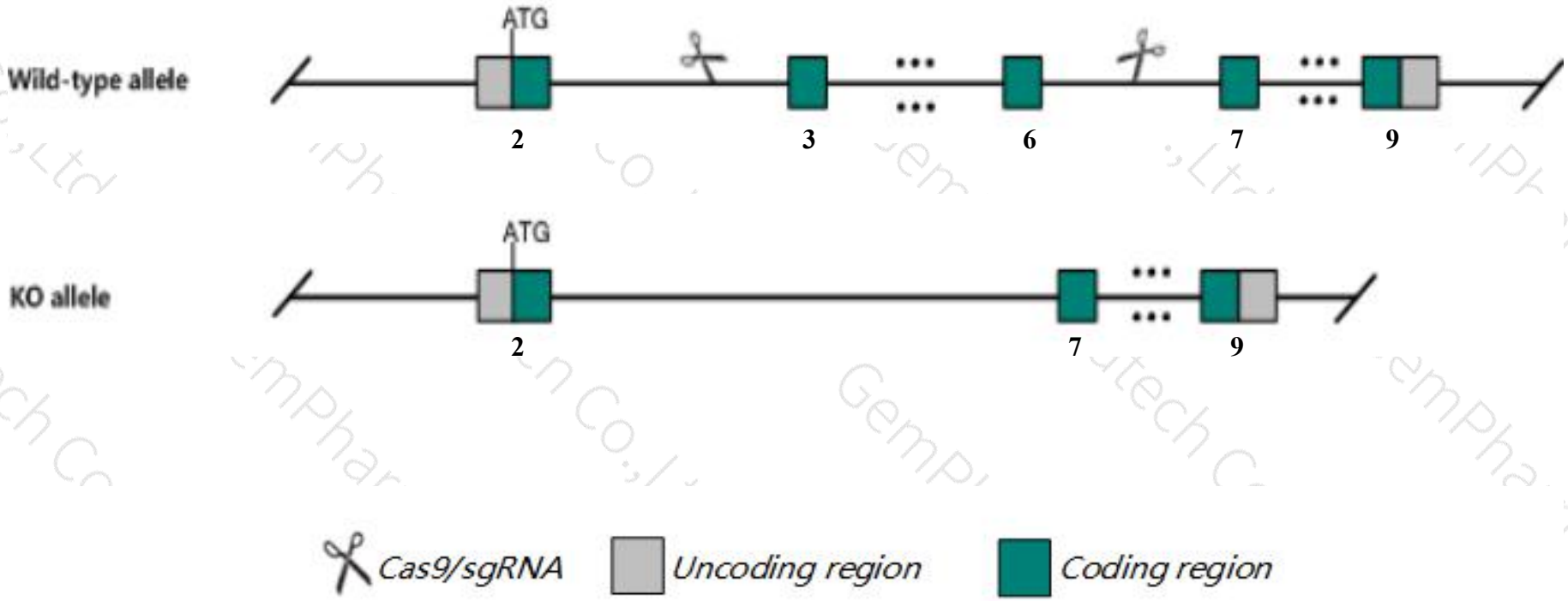
Design Date: 2021-1-8

Project Overview

Project Name	***Syt2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Syt2* gene. The schematic diagram is as follows:



Technical routes

➢ The *Syt2* gene has 4 transcripts. According to the structure of *Syt2* gene, exon3-exon6 of *Syt2-201*(ENSMUST00000121990.1) transcript is recommended as the knockout region.The region contains 623bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Syt2* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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

➢ According to the existing MGI data,mice homozygous for an ENU-induced allele are viable but sterile, weigh less and show ataxia and altered spontaneous and Ca2+-evoked neurotransmitter release. Mice homozygous for a null allele die at weaning showing growth arrest, motor dysfunction and impaired Ca2+-evoked neurotransmitter release.

➢ The *Syt2* gene is located on the Chr1. 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）

集萃药康 GemPharmatech

Syt2 synaptotagmin II [Mus musculus (house mouse)]

Gene ID: 20980, updated on 13-Mar-2020

Summary

Official Symbol	Syt2 provided by MGI
Official Full Name	synaptotagmin II provided byMGI
Primary source	MGI:MGI:99666
See related	Ensembl:ENSMUSG00000026452
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	IP4BP, R74640, mKIAA4194, sytII
Summary	This gene encodes a member of the synaptotagmin protein family. Synaptotagmin proteins are involved in membrane trafficking and are characterized by an N-terminal transmembrane region as well as tandem calcium binding domains. The encoded protein is able to bind inositol polyphosphate and is thought to be involved in synaptic function and neurotransmitter release. [provided by RefSeq, Sep 2017]
Expression	Biased expression in cerebellum adult (RPKM 41.6) and cortex adult (RPKM 3.9)See more
Orthologs	human all

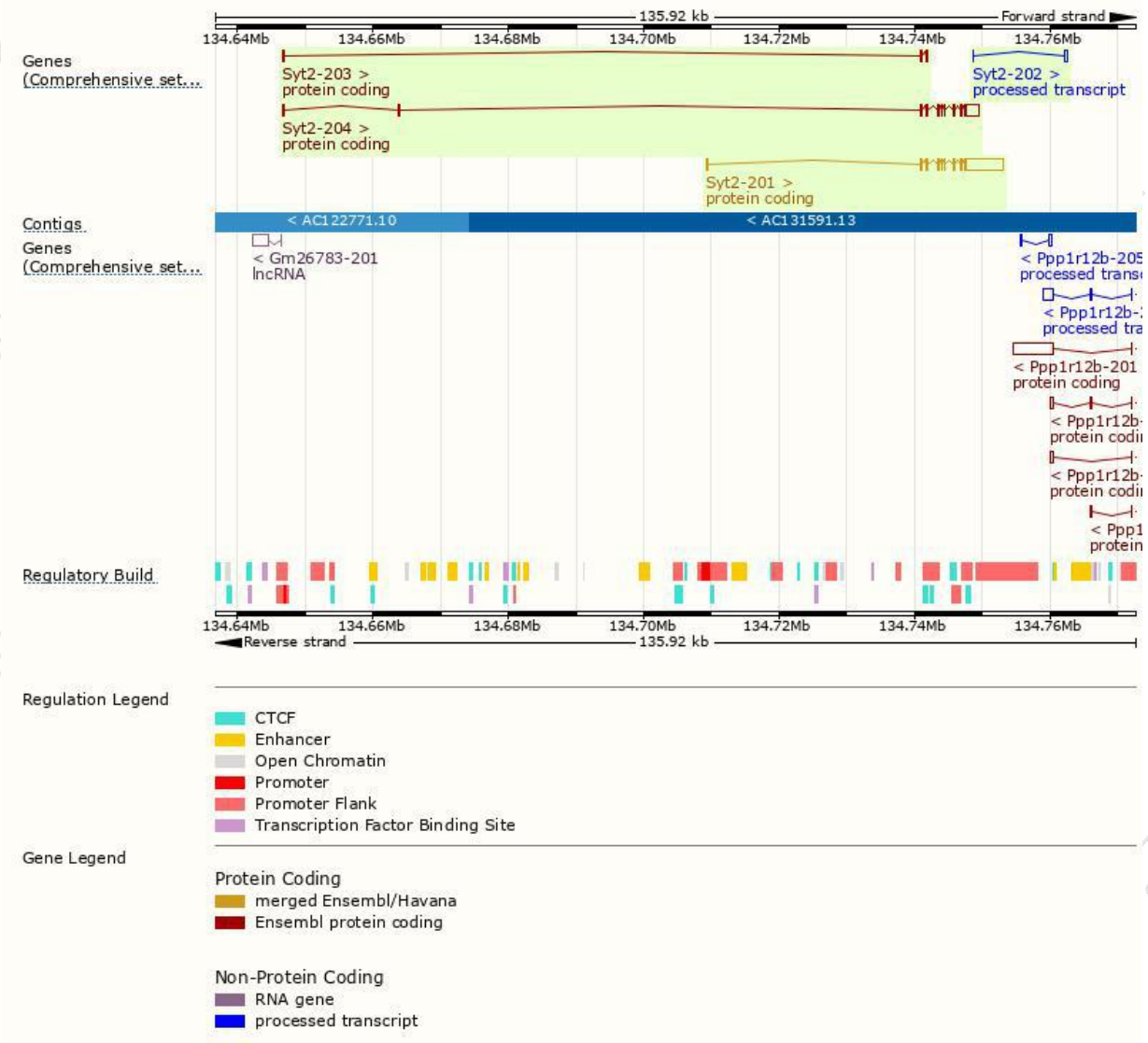
Transcript information（Ensembl）

The gene has 4 transcripts,all transcripts are shown below:

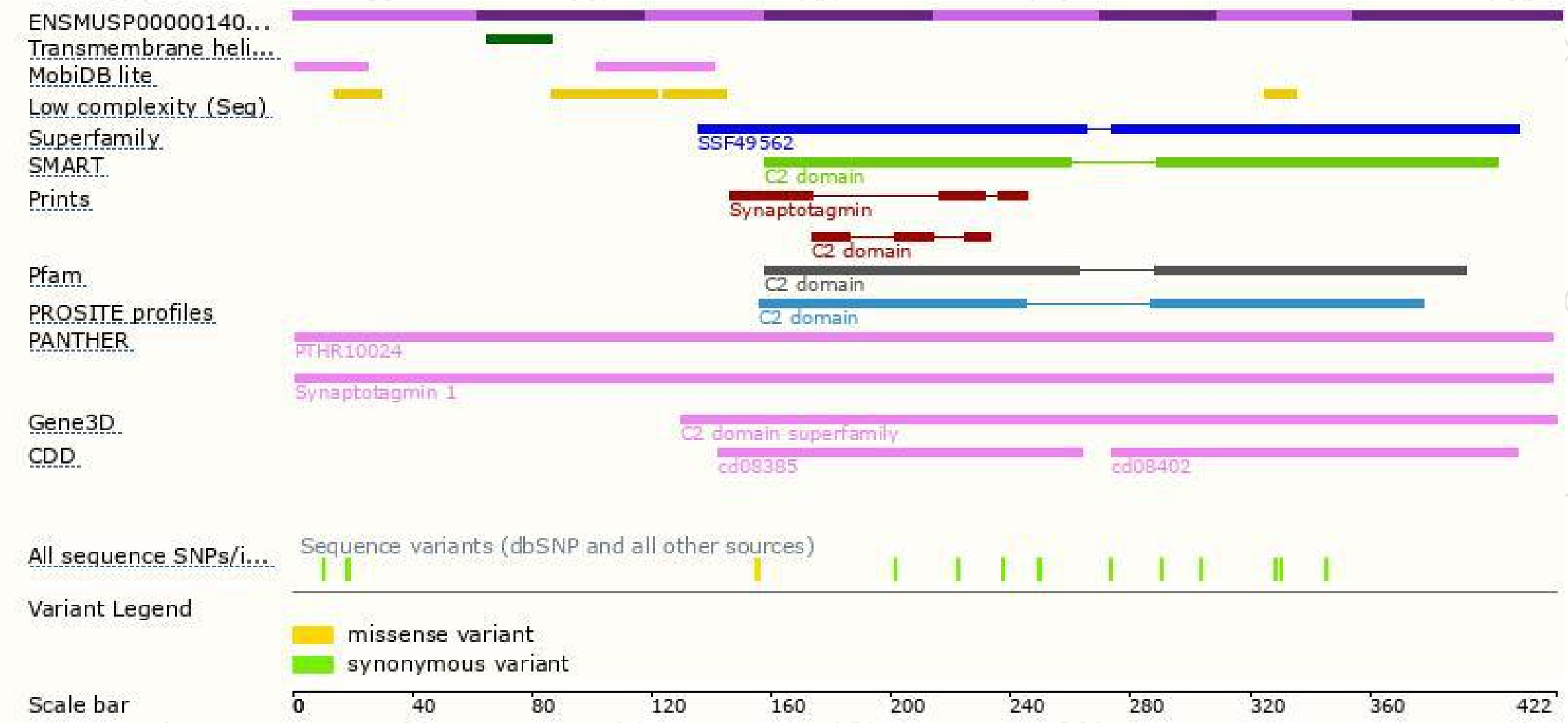
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Syt2-201	ENSMUST00000121990.1	6879	422aa	Protein coding	CCDS15312	A0A0R4J2C2	TSL:1 GENCODE basic APPRIS P1
Syt2-204	ENSMUST00000188842.6	3386	422aa	Protein coding	CCDS15312	A0A0R4J2C2	TSL:1 GENCODE basic APPRIS P1
Syt2-203	ENSMUST00000187725.6	383	69aa	Protein coding	-	Q9JM87	CDS 3' incomplete TSL:1
Syt2-202	ENSMUST00000187702.1	485	No protein	Processed transcript	-	-	TSL:3

The strategy is based on the design of *Syt2-201* transcript,the transcription is shown below:

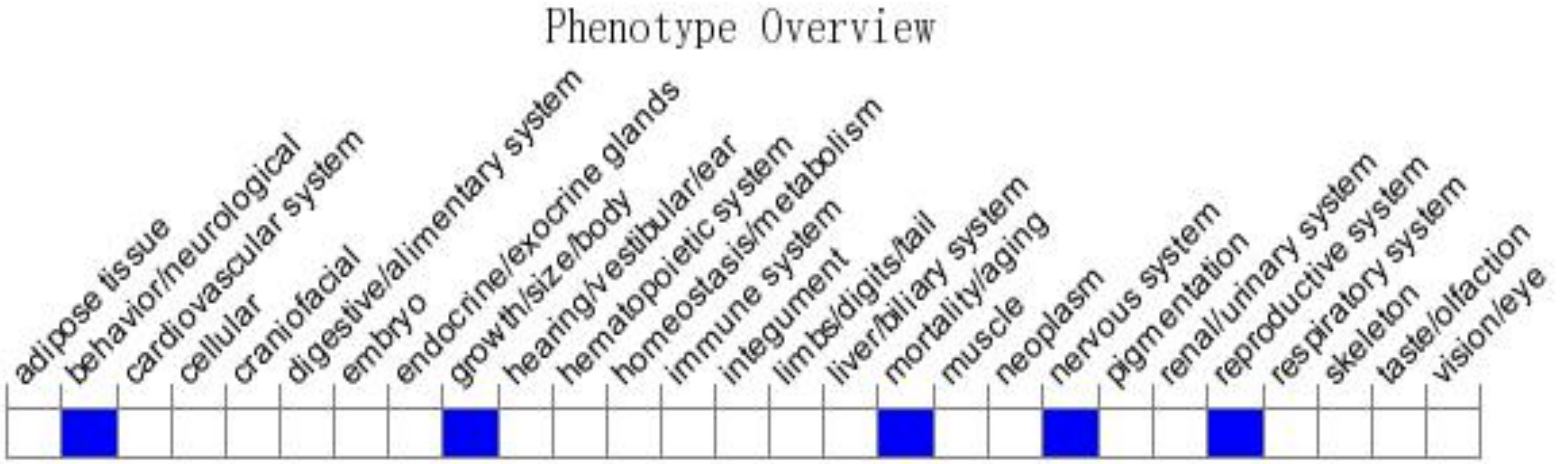
Genomic location distribution



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 an ENU-induced allele are viable but sterile, weigh less and show ataxia and altered spontaneous and Ca2+-evoked neurotransmitter release. Mice homozygous for a null allele die at weaning showing growth arrest, motor dysfunction and impaired Ca2+-evoked neurotransmitter release.

集萃药康
GemPharmatech

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

Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd