

# Slc46a1 Cas9-CKO Strategy

Designer:Xueting Zhang

Reviewer: Yanhua Shen

Date:2020-02-12

## **Project Overview**



**Project Name** 

Slc46a1

**Project type** 

Cas9-CKO

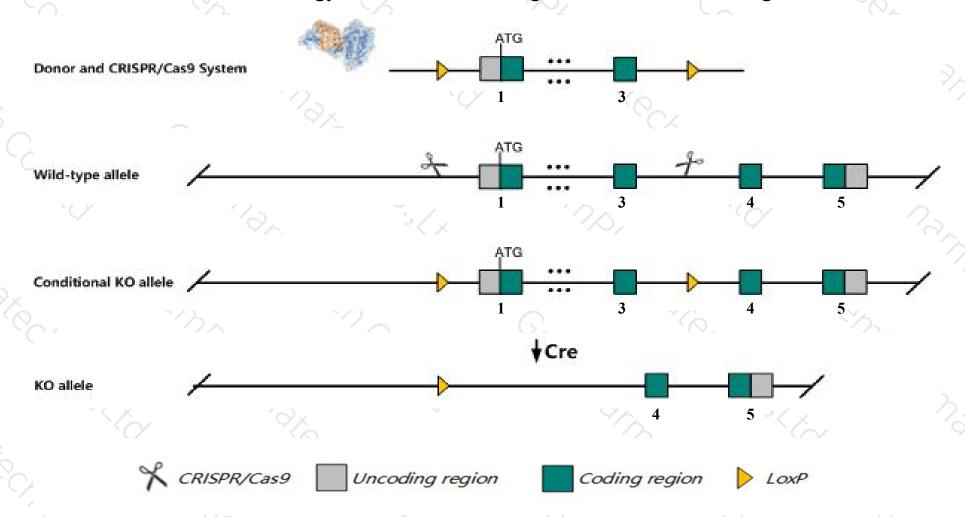
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Slc46a1 gene. The schematic diagram is as follows:



### Technical routes



- The *Slc46a1* gene has 3 transcripts. According to the structure of *Slc46a1* gene, exon1-exon3 of *Slc46a1-201* (ENSMUST0000001126.3) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Slc46a1* 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 exhibit increased circulating and liver levels of N-homocysteine and total homocysteine.
- > Transcript *Slc46a1*-203 may not be affected.
- The *Slc46a1* gene is located on the Chr11. 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)



#### SIc46a1 solute carrier family 46, member 1 [ Mus musculus (house mouse) ]

Gene ID: 52466, updated on 12-Aug-2019

#### Summary

☆ ?

Official Symbol Slc46a1 provided by MGI

Official Full Name solute carrier family 46, member 1 provided by MGI

Primary source MGI:MGI:1098733

See related Ensembl: ENSMUSG00000020829

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 HCP1; Pcft; D11Ertd18e; 1110002C08Rik

**Expression** Biased expression in duodenum adult (RPKM 274.9), small intestine adult (RPKM 108.1) and 8 other tissues See more

Orthologs human all

#### Genomic context



**Location:** 11 B5; 11 46.74 cM

See Slc46a1 in Genome Data Viewer

Exon count: 5

Annotation release	Status	Assembly	Chr	hr Location	
<u>108</u>	current	GRCm38.p6 (GCF_000001635.26)	11	NC_000077.6 (7846570178471945)	
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	11	NC_000077.5 (7827920378285447)	

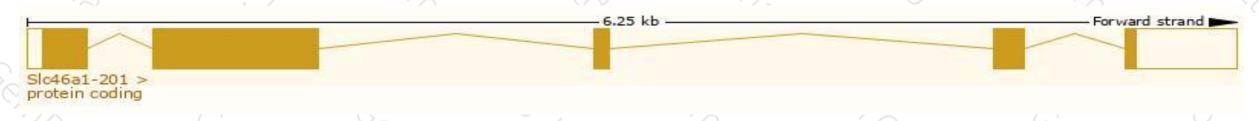
## Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

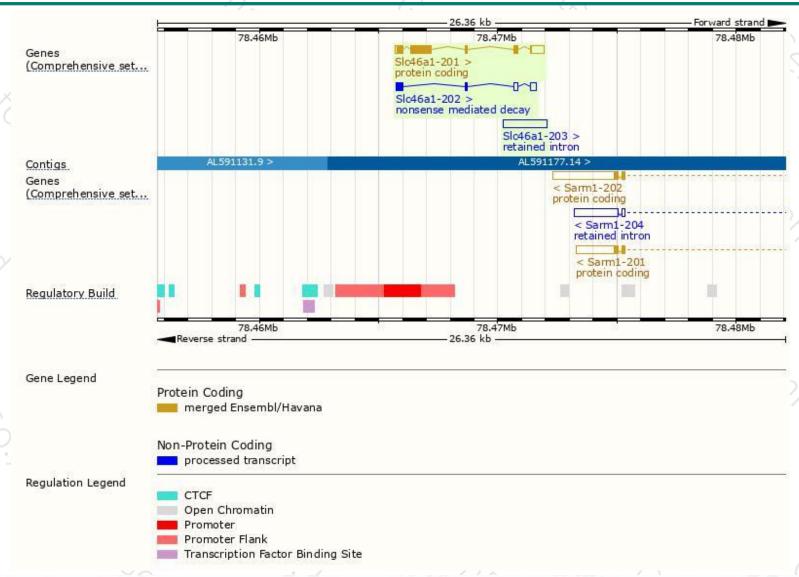
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
SIc46a1-201	ENSMUST00000001126.3	1978	459aa	Protein coding	CCDS25104	Q6PEM8	TSL:1 GENCODE basic APPRIS P1
SIc46a1-202	ENSMUST00000146431.2	749	<u>98aa</u>	Nonsense mediated decay	8 <del>-</del>	F6QD87	TSL:3
SIc46a1-203	ENSMUST00000180786.1	1834	No protein	Retained intron	¥4	ů.	TSL:NA

The strategy is based on the design of Slc46a1-201 transcript, The transcription is shown below



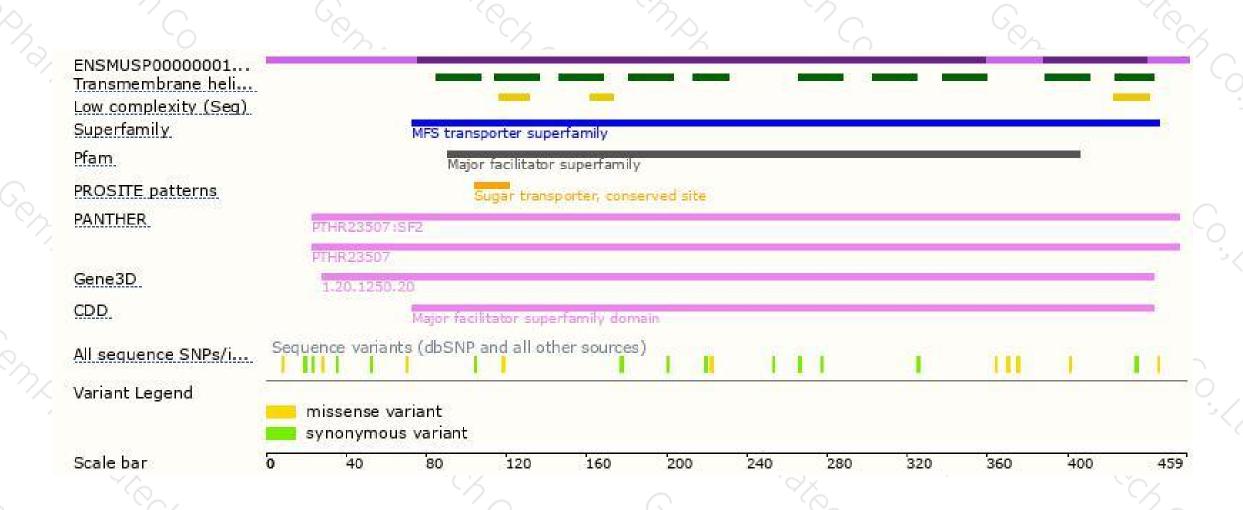
### Genomic location distribution





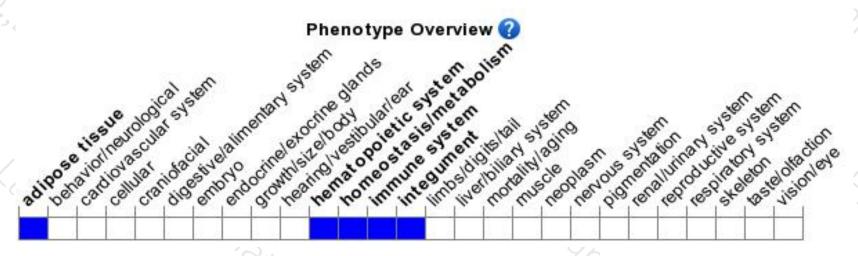
### Protein domain





## 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 exhibit increased circulating and liver levels of N-homocysteine and total homocysteine.



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





