# *Slc22a14* Cas9-CKO Strategy

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**Reviewer: Xiaojing Li**

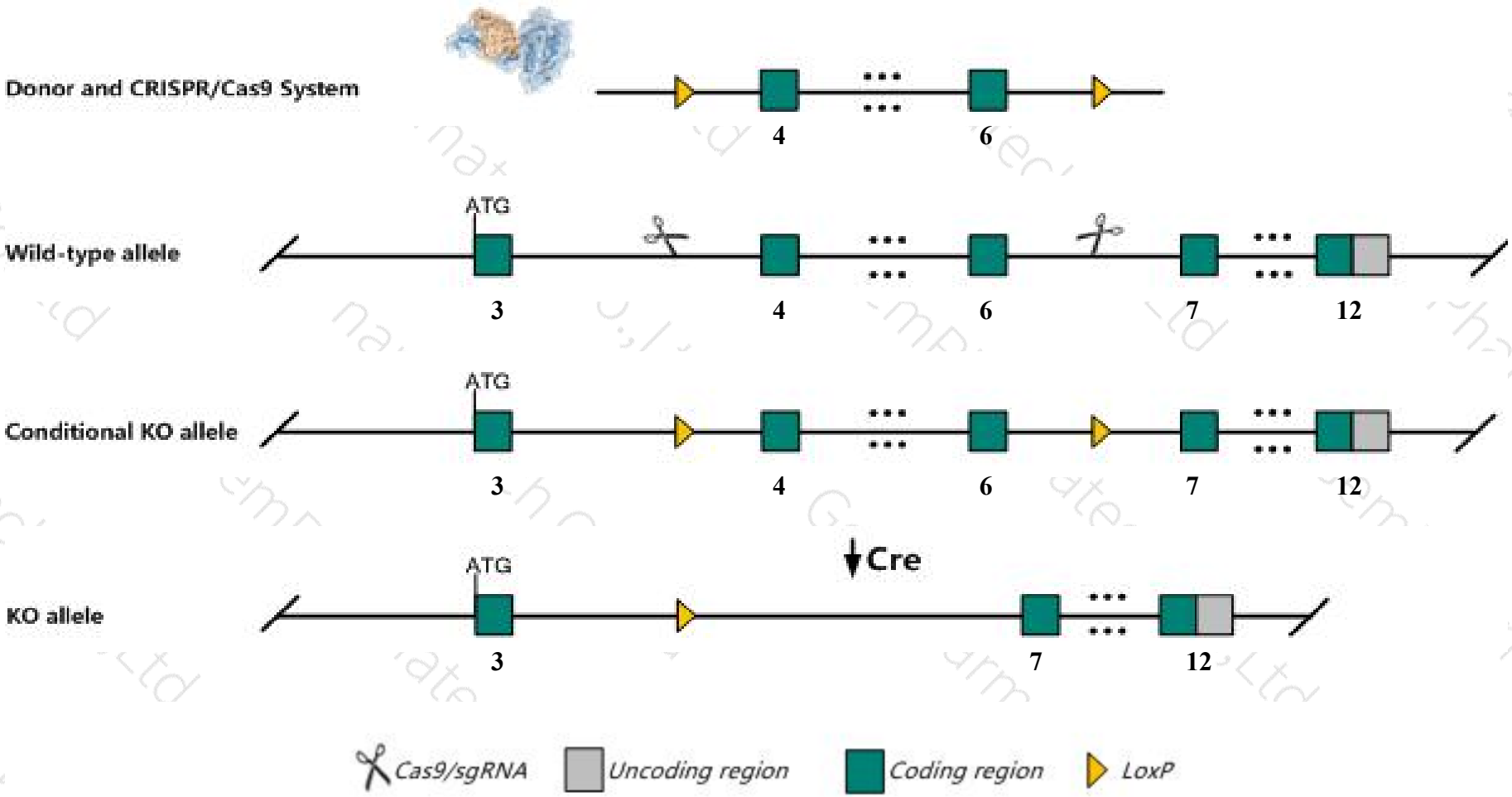
**Design Date: 2020-9-16**

# Project Overview

| | |
|---|---|
| **Project Name** | ***Slc22a14*** |
| **Project type** | **Cas9-CKO** |
| **Strain background** | **C57BL/6JGpt** |

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Slc22a14* gene. The schematic diagram is as follows:



# Technical routes

➢ The *Slc22a14* gene has 4 transcripts. According to the structure of *Slc22a14* gene, exon4-exon6 of *Slc22a14-201*(ENSMUST00000093775.11) transcript is recommended as the knockout region.The region contains 428bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Slc22a14* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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 severe male infertility associated with asthenozoospermia, impaired sperm capacitation, decreased fertilization frequency, abnormal sperm flagellar bending, and abnormal sperm annulus morphology.

➢ Transcript *Slc22a14*-202, *Slc22a14*-203 may not be affected.

➢ The *Slc22a14* gene is located on the Chr9. 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）

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## Slc22a14 solute carrier family 22 (organic cation transporter), member 14 [Mus musculus (house mouse)]

Gene ID: 382113, updated on 13-Mar-2020

### Summary

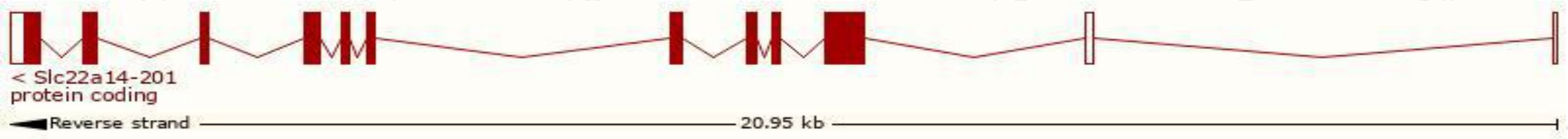
| | |
|---|---|
| **Official Symbol** | Slc22a14 provided by MGI |
| **Official Full Name** | solute carrier family 22 (organic cation transporter), member 14 provided byMGI |
| **Primary source** | MGI:MGI:2685974 |
| **See related** | Ensembl:ENSMUSG00000070280 |
| **Gene type** | protein coding |
| **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** | Gm1128 |
| **Expression** | Restricted expression toward testis adult (RPKM 114.3)See more |
| **Orthologs** | human all |

# Transcript information（Ensembl）

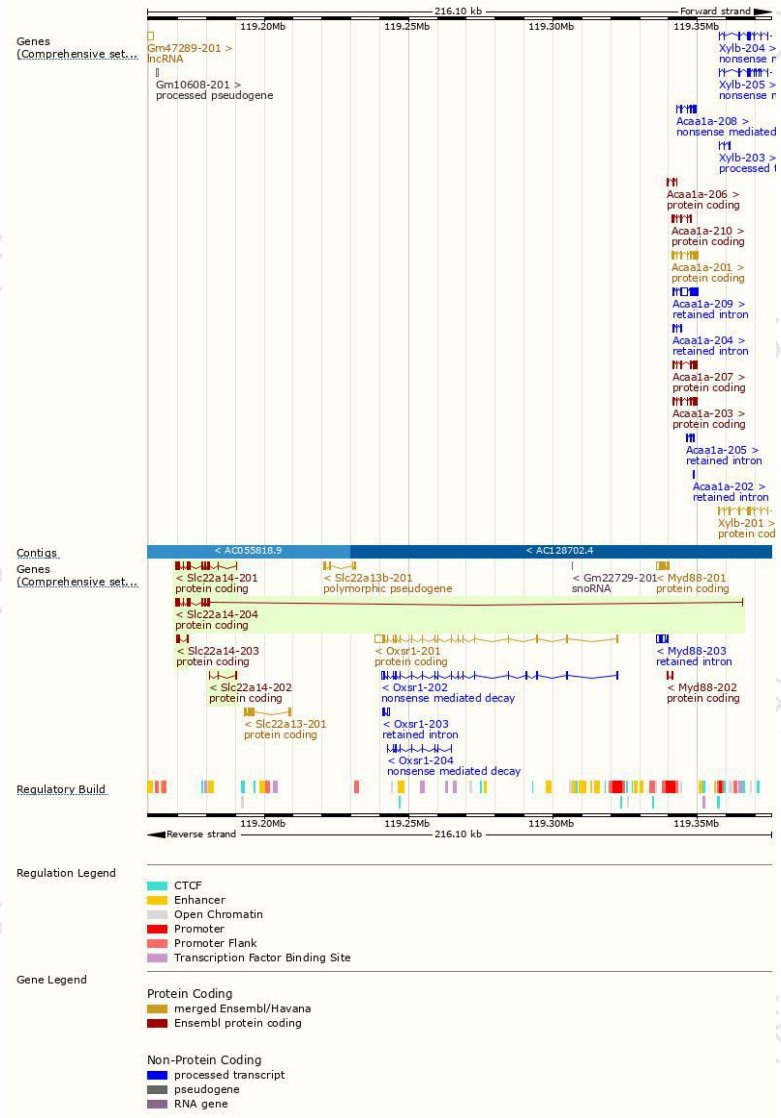
The gene has 4 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| **Slc22a14-201** | ENSMUST00000093775.11 | 2213 | 629aa | Protein coding | CCDS23609 | Q497L9 | TSL:1 GENCODE basic APPRIS P1 |
| **Slc22a14-204** | ENSMUST00000170400.8 | 2110 | 629aa | Protein coding | CCDS23609 | Q497L9 | TSL:5 GENCODE basic APPRIS P1 |
| **Slc22a14-203** | ENSMUST00000152061.1 | 589 | 196aa | Protein coding | - | F7AMC9 | CDS 5' and 3' incomplete TSL:3 |
| **Slc22a14-202** | ENSMUST00000127794.1 | 340 | 20aa | Protein coding | - | D3YUH1 | CDS 3' incomplete TSL:5 |

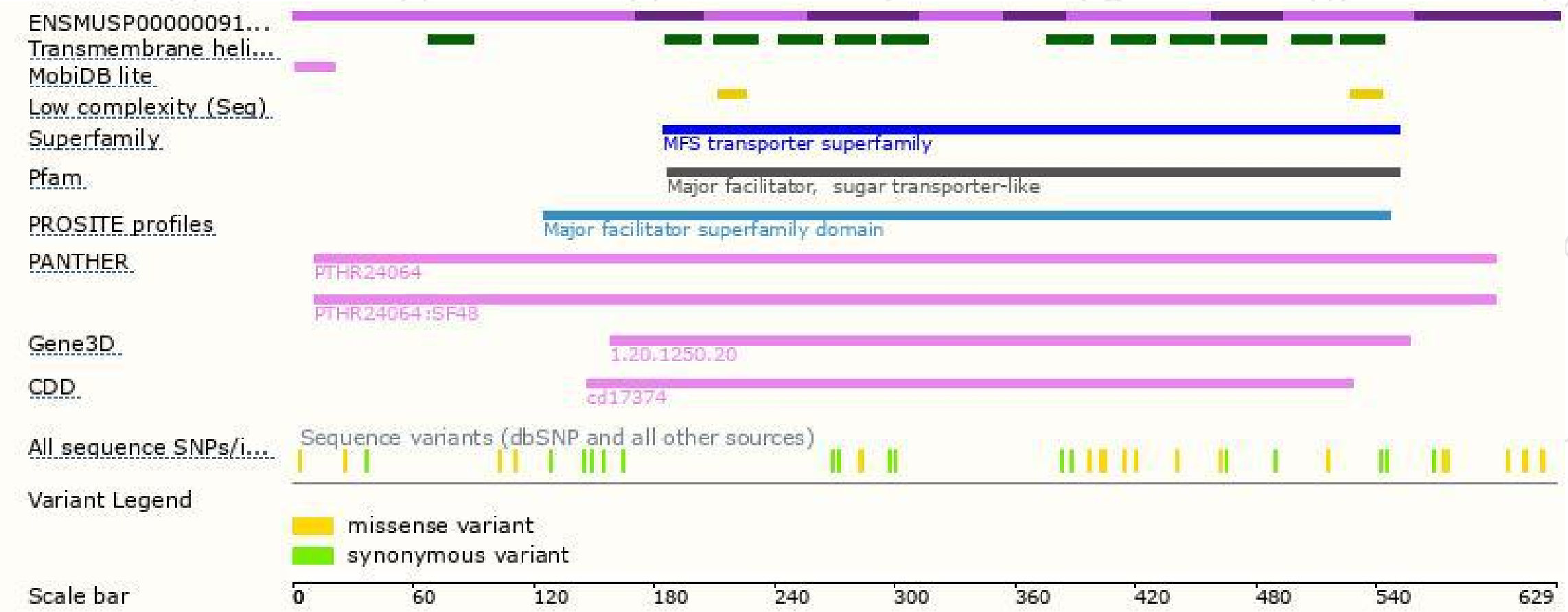
The strategy is based on the design of *Slc22a14-201* transcript,the transcription is shown below:



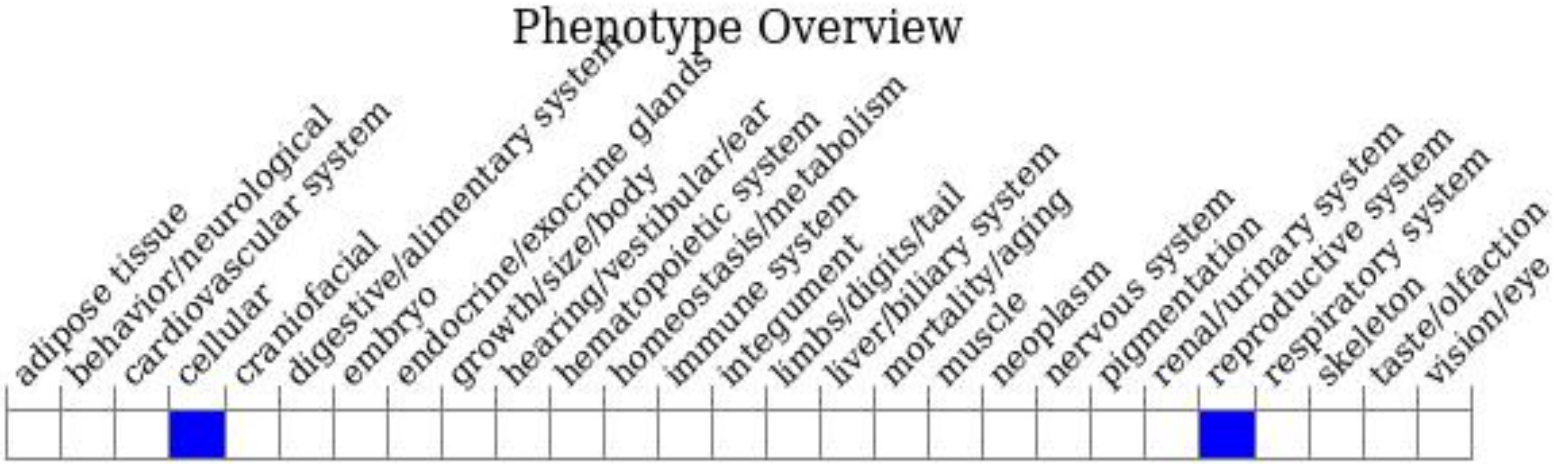
# Genomic location distribution



# 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 severe male infertility associated with asthenozoospermia, impaired sperm capacitation, decreased fertilization frequency, abnormal sperm flagellar bending, and abnormal sperm annulus morphology.

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

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