# *Cldn7* Cas9-CKO Strategy

**Designer:** Huan Wang
**Reviewer:** Huan Fan

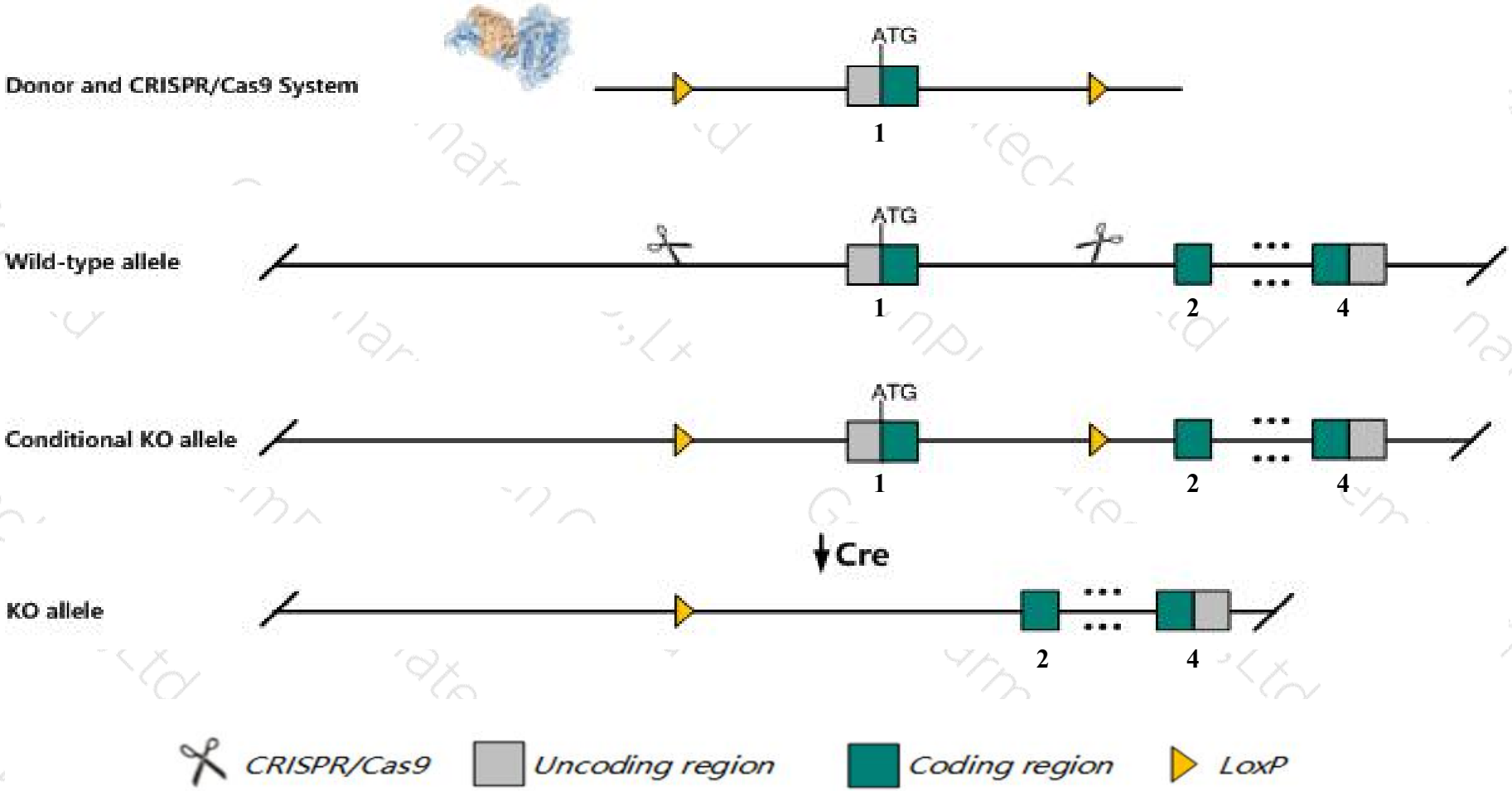
**Design Date:** 2020-1-4

# Project Overview

| | |
|---|---|
| **Project Name** | ***Cldn7*** |
| **Project type** | **Cas9-CKO** |
| **Strain background** | **C57BL/6JGpt** |

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cldn7* gene. The schematic diagram is as follows:



# Technical routes

- The *Cldn7* gene has 5 transcripts. According to the structure of *Cldn7* gene, exon1 of *Cldn7-201* (ENSMUST00000018713.12) 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 *Cldn7* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 decreased body size, weight, and length; abnormal potassium, chloride, and sodium ion excretion; chronic dehydration; and postnatal lethality by P12.
- Transcript *Cldn7-205* may not be affected.

- The *Cldn7* 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）

集萃药康 GemPharmatech

## Cldn7 claudin 7 [Mus musculus (house mouse)]

Gene ID: 53624, updated on 9-Apr-2019

### Summary

| | |
|---|---|
| **Official Symbol** | Cldn7 provided by MGI |
| **Official Full Name** | claudin 7 provided byMGI |
| **Primary source** | MGI:MGI:1859285 |
| **See related** | Ensembl:ENSMUSG00000018569 |
| **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 |
| **Summary** | This gene encodes a member of the claudin family. Claudins are integral membrane proteins and components of tight junction strands. Tight junction strands serve as a physical barrier to prevent solutes and water from passing freely through the paracellular space between epithelial or endothelial cell sheets, and also play critical roles in maintaining cell polarity and signal transductions. This gene is expressed constitutively in the mammary epithelium throughout development, and might be involved in vesicle trafficking to the basolateral membrane. It is essential for NaCl homeostasis in distal nephrons. The knockout mice lacking this gene showed severe salt wasting, chronic dehydration, and growth retardation, and died within 12 days after birth. Alternatively spliced transcript variants have been found for this gene. [provided by RefSeq, Aug 2010] |
| **Expression** | Biased expression in large intestine adult (RPKM 431.6), small intestine adult (RPKM 310.8) and 4 other tissuesSee more |
| **Orthologs** | human all |

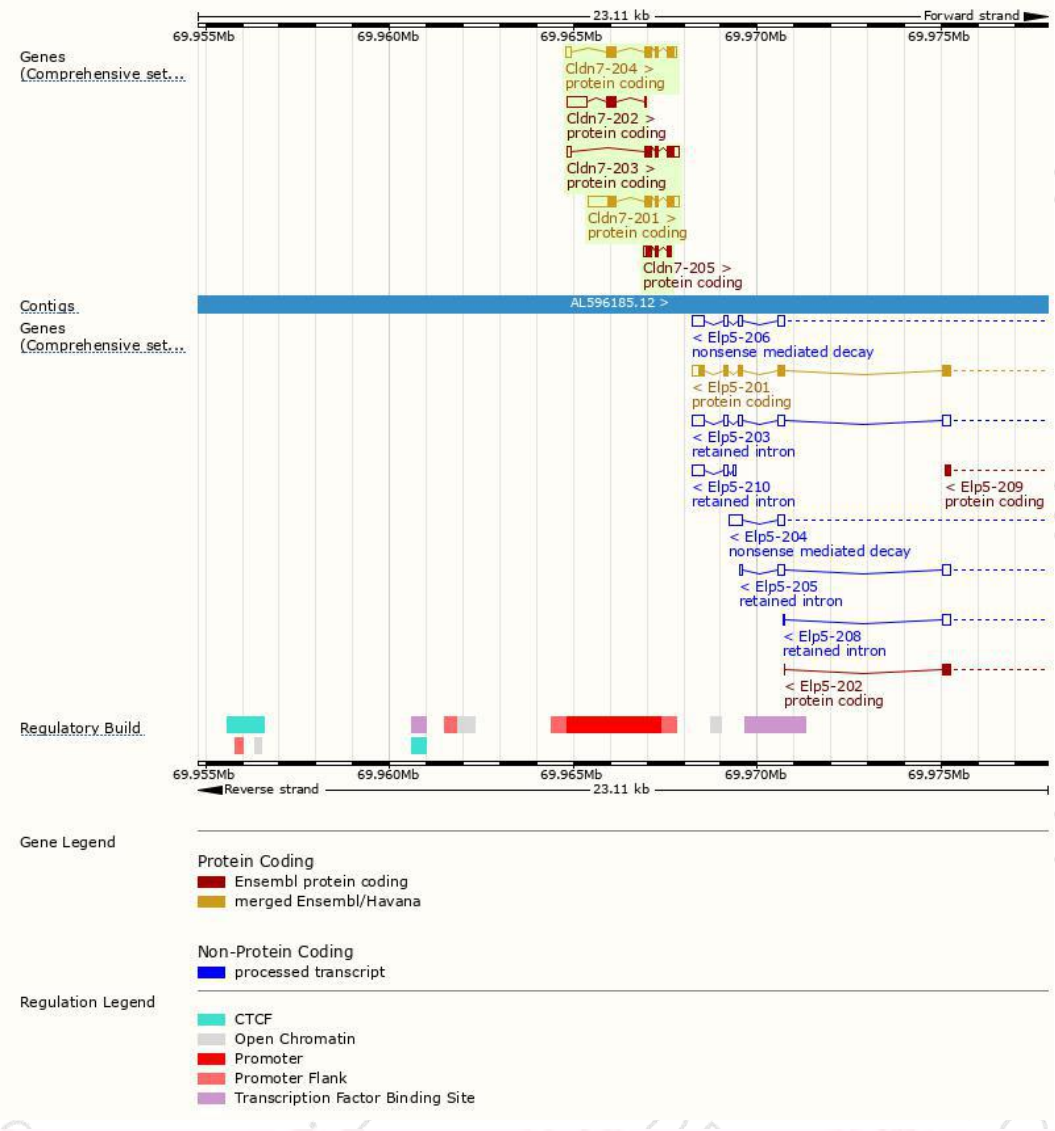
# Transcript information（Ensembl）

The gene has 5 transcripts,all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| Cldn7-201 | ENSMUST00000018713.12 | 1335 | 211aa | Protein coding | CCDS24925 | Q9Z261 | TSL:1 GENCODE basic APPRIS P1 |
| Cldn7-204 | ENSMUST00000108597.7 | 913 | 211aa | Protein coding | CCDS24925 | Q9Z261 | TSL:2 GENCODE basic APPRIS P1 |
| Cldn7-202 | ENSMUST00000060651.5 | 835 | 83aa | Protein coding | - | B1AR46 | CDS 3' incomplete TSL:5 |
| Cldn7-203 | ENSMUST00000108596.7 | 664 | 128aa | Protein coding | - | B1AR47 | TSL:3 GENCODE basic |
| Cldn7-205 | ENSMUST00000151515.1 | 373 | 107aa | Protein coding | - | J3QQ48 | CDS 3' incomplete TSL:2 |

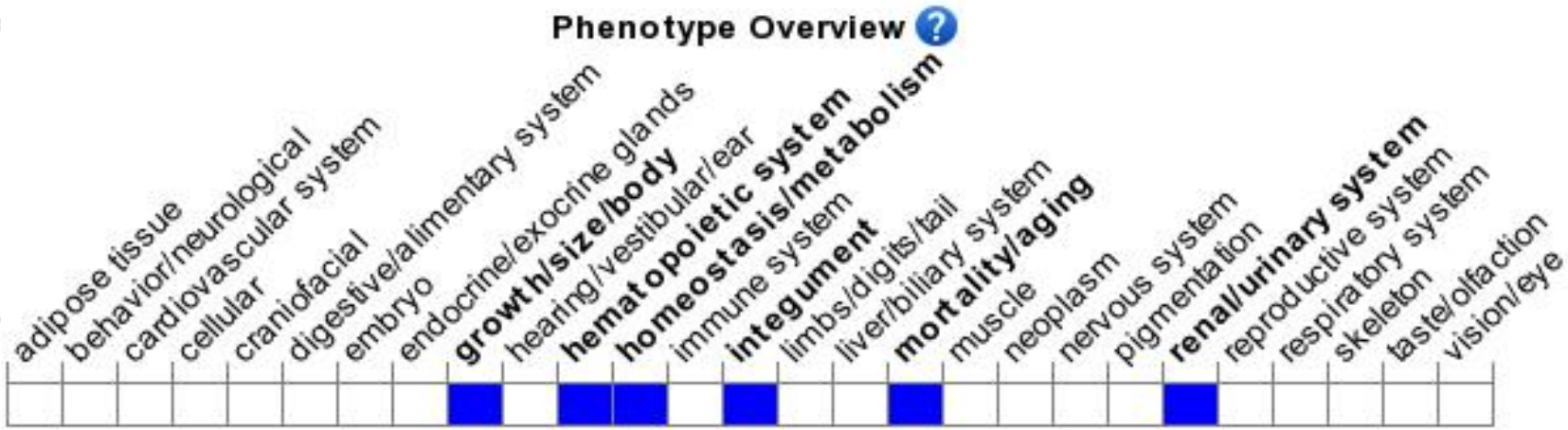
The strategy is based on the design of *Cldn7-201* transcript,The transcription is shown below

# 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 decreased body size, weight, and length; abnormal potassium, chloride, and sodium ion excretion; chronic dehydration; and postnatal lethality by P12.

集萃药康
GemPharmatech

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

Tel: 400-9660890

集萃药康生物科技
GemPharmatech Co.,Ltd