

# Celsr3 Cas9-CKO Strategy

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**Reviewer:** 

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**Design Date:** 

2018-6-28

# **Project Overview**



**Project Name** 

Celsr3

**Project type** 

Cas9-CKO

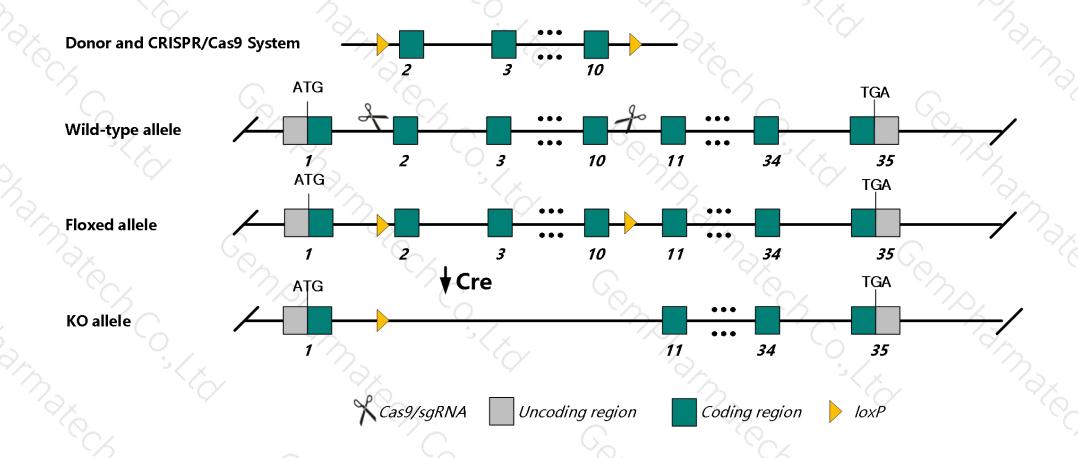
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Celsr3 gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Celsr3* gene has 7 transcripts. According to the structure of *Celsr3* gene, exon2-exon10 of *Celsr3-201*(ENSMUST00000024238.10) transcript is recommended as the knockout region. The region contains 1880bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Celsr3* 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 null allele exhibit neonatal lethality, abnormal neurvous system development, and abnormal respiratory system development.
- ➤ Transcript Celsr3-204/205/206 may not be affected. The size of intron10 for 3'-loxP site insertion is 558bp.

  The KO region is close to 5'UTR region of the Gm35025 gene. Knockout the region may affect the regulatory function of Gm35025 gene.
- The *Celsr3* 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)



#### Celsr3 cadherin, EGF LAG seven-pass G-type receptor 3 [ Mus musculus (house mouse) ]

Gene ID: 107934, updated on 14-Aug-2019

#### Summary



Official Symbol Celsr3 provided by MGI

Official Full Name cadherin, EGF LAG seven-pass G-type receptor 3 provided by MGI

Primary source MGI:MGI:1858236

See related Ensembl: ENSMUSG00000023473

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 Fmi1; Adgrc3; flamingo; mKIAA0812

Expression Biased expression in whole brain E14.5 (RPKM 20.1), CNS E18 (RPKM 16.6) and 6 other tissues See more

Orthologs human all

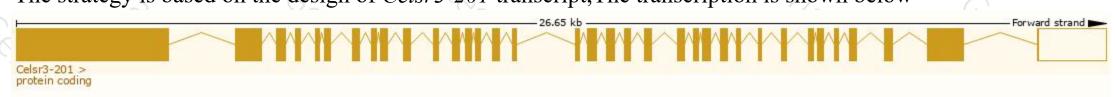
# Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

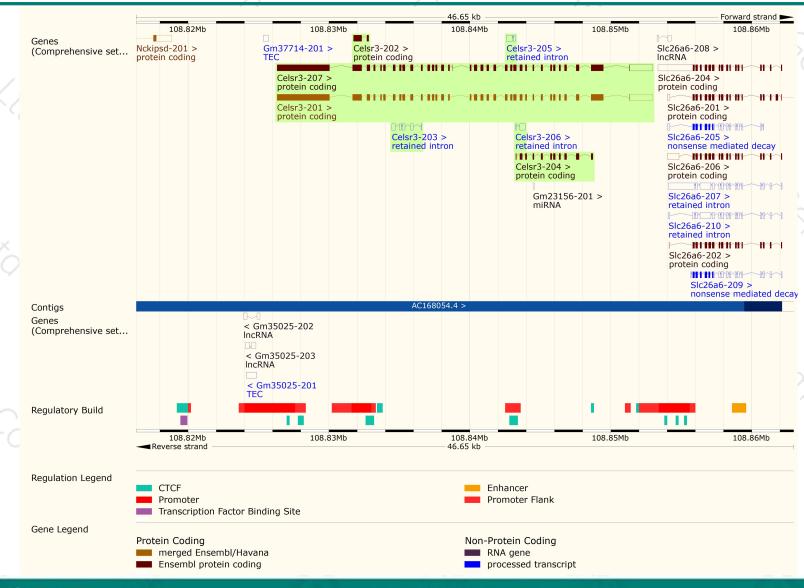
Name A	Transcript ID	bp 🍦	Protein	Translation ID 🔷	Biotype	CCDS 🍦	UniProt	Flags
Celsr3-201	ENSMUST00000024238.10	11547	3301aa	ENSMUSP00000024238.5	Protein coding	2	Q91ZI0 &	TSL:1 GENCODE basic APPRIS ALT2
Celsr3-202	ENSMUST00000192235.1	722	240aa	ENSMUSP00000141429.1	Protein coding	=	<u>A0A0A6YW76</u> ₽	CDS 5' and 3' incomplete TSL:3
Celsr3-203	ENSMUST00000193726.1	767	No protein	-	Retained intron	-	-	TSL:5
Celsr3-204	ENSMUST00000194079.1	1418	472aa	ENSMUSP00000141789.1	Protein coding	-	A0A0A6YX12 ₺	CDS 5' and 3' incomplete TSL:1
Celsr3-205	ENSMUST00000195255.1	524	No protein	(#)	Retained intron	=	(0)	TSL:2
Celsr3-206	ENSMUST00000195857.1	443	No protein	650	Retained intron	5.	3 <u>=</u> 1	TSL:2
Celsr3-207	ENSMUST00000213524.2	11571	3309aa	ENSMUSP00000150759.1	Protein coding	3	A0A1L1SUG9@	TSL:1   GENCODE basic   APPRIS P5

The strategy is based on the design of Celsr3-201 transcript, The transcription is shown below



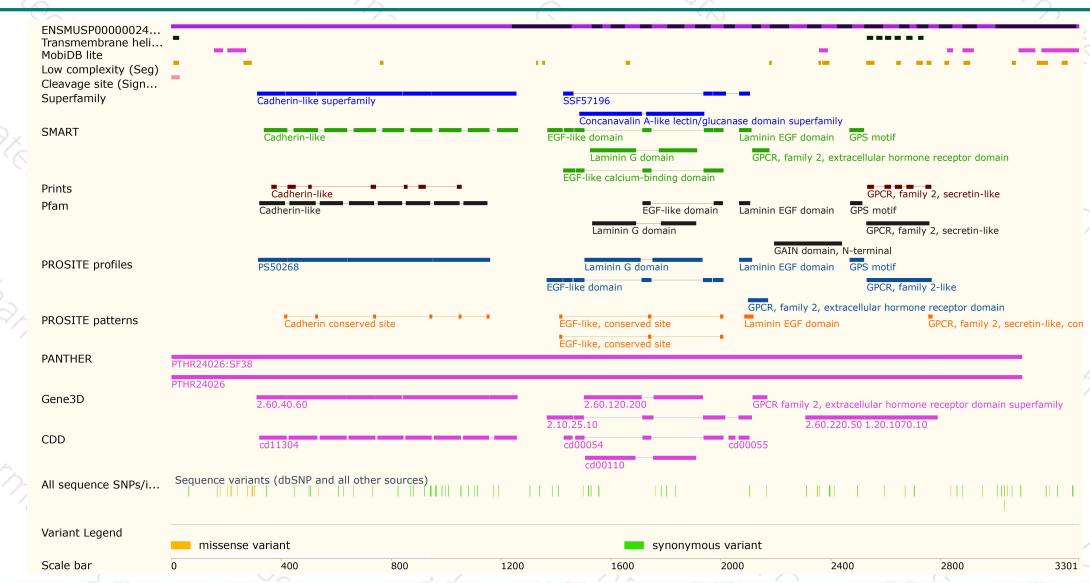
### Genomic location distribution





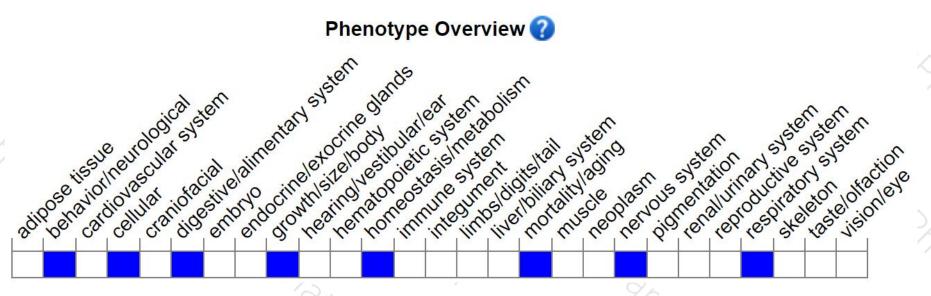
### Protein domain





# 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 null allele exhibit neonatal lethality, abnormal neurvous system development, and abnormal respiratory system development.



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





