

# Gmcl1 Cas9-CKO Strategy

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Reviewer: Xiaojing Li

**Design Date: 2020-8-3** 

# **Project Overview**



**Project Name** 

Gmcl1

**Project type** 

Cas9-CKO

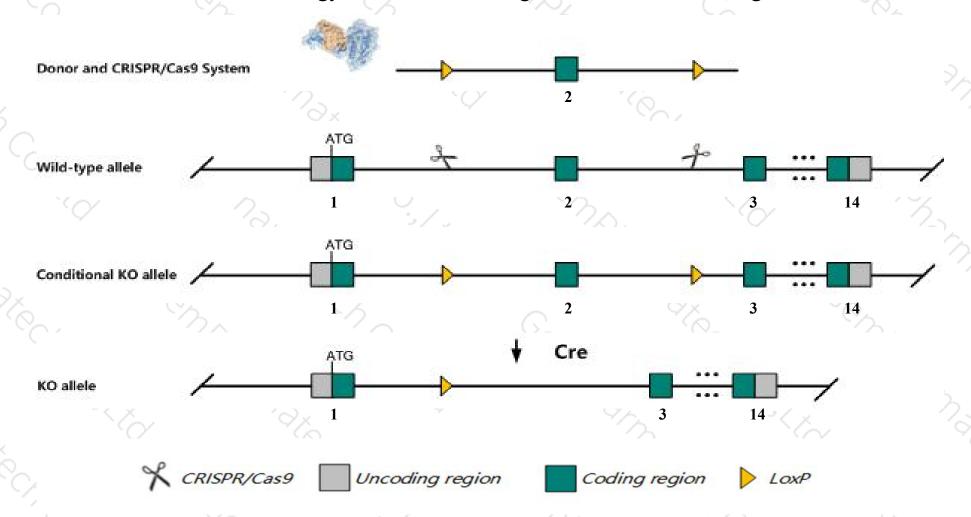
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Gmcl1* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Gmcl1* gene has 4 transcripts. According to the structure of *Gmcl1* gene, exon2 of *Gmcl1*201(ENSMUST00000001185.13) transcript is recommended as the knockout region. The region contains 124bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Gmcl1* 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, homozygotes for a targeted null mutation exhibit impaired nuclear membrane integrity in liver, endocrine pancreas, and testis. Mutant males show reduced fertility with decreased chromatin condensation and morphologically abnormal sperm.
- > The *Gmcl1* gene is located on the Chr6. 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)



#### Gmcl1 germ cell-less, spermatogenesis associated 1 [Mus musculus (house mouse)]

Gene ID: 23885, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Gmcl1 provided by MGI

Official Full Name germ cell-less, spermatogenesis associated 1 provided by MGI

Primary source MGI:MGI:1345156

See related Ensembl:ENSMUSG00000001157

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 2810049L19Rik, Btbd13, DIP, Gcl, Gcl-1, glc-1, mglc-1

Expression Ubiquitous expression in testis adult (RPKM 18.2), CNS E14 (RPKM 8.7) and 27 other tissuesSee more

Orthologs human all

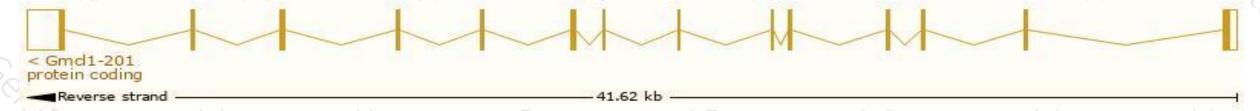
# Transcript information (Ensembl)



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

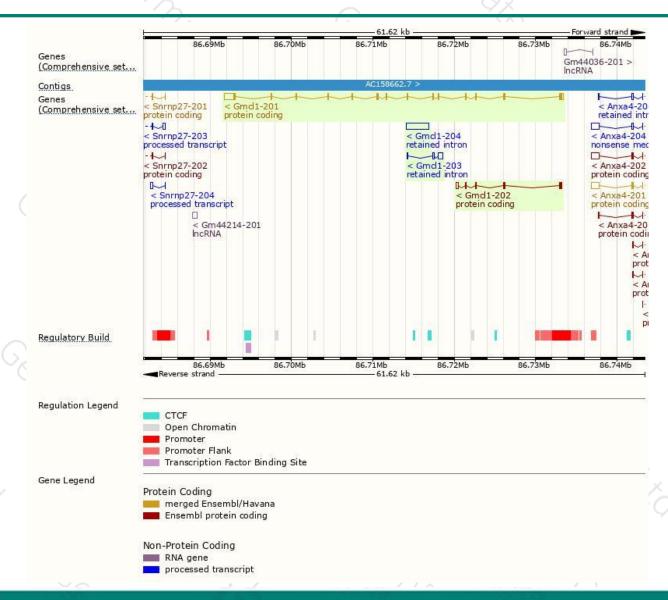
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Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gmcl1-201	ENSMUST00000001185.13	2949	<u>524aa</u>	Protein coding	CCDS20316	Q920G9	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Gmcl1-202	ENSMUST00000113679.1	960	<u>199aa</u>	Protein coding	(-)	Q3V0Z2	TSL:1 GENCODE basic
Gmcl1-204	ENSMUST00000203741.1	2750	No protein	Retained intron	923	-	TSL:NA
Gmcl1-203	ENSMUST00000135880.1	779	No protein	Retained intron	-	12	TSL:3

The strategy is based on the design of *Gmcl1-201* transcript, the transcription is shown below:



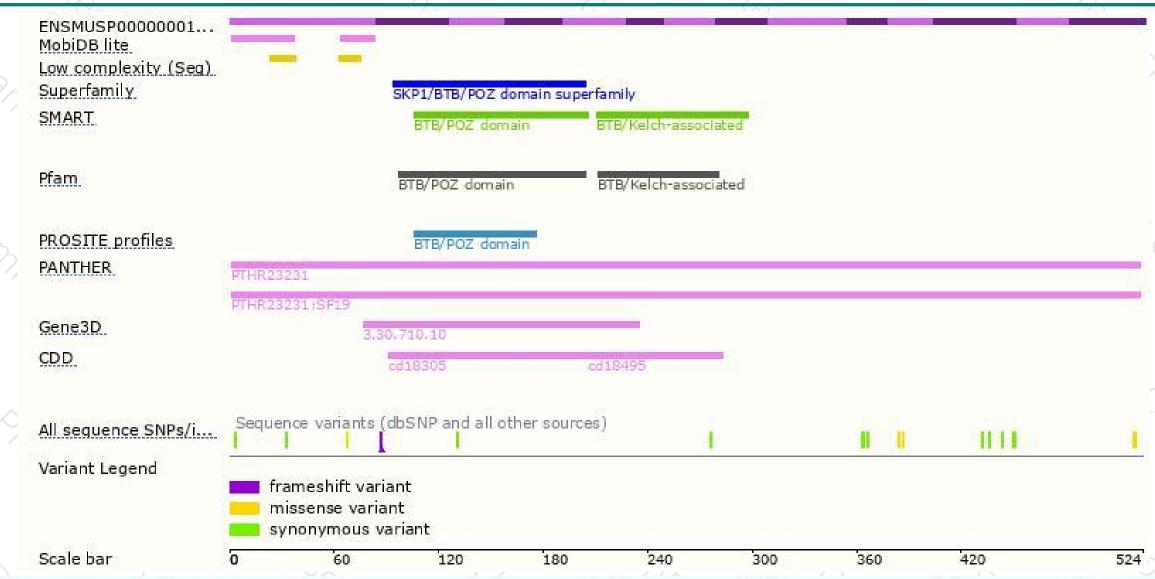
### Genomic location distribution





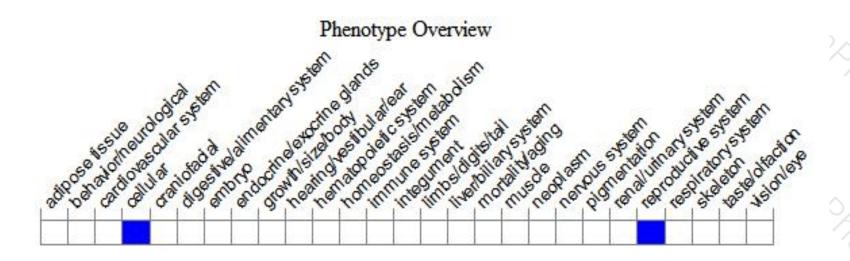
### Protein domain





# 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, homozygotes for a targeted null mutation exhibit impaired nuclear membrane integrity in liver, endocrine pancreas, and testis. Mutant males show reduced fertility with decreased chromatin condensation and morphologically abnormal sperm.



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





