

# *Cytip* Cas9-CKO Strategy

Designer: Huan Fan

Reviewer: Huan Wang

Design Date: 2020-5-26

# Project Overview

---

**Project Name**

*Cytip*

---

**Project type**

**Cas9-CKO**

---

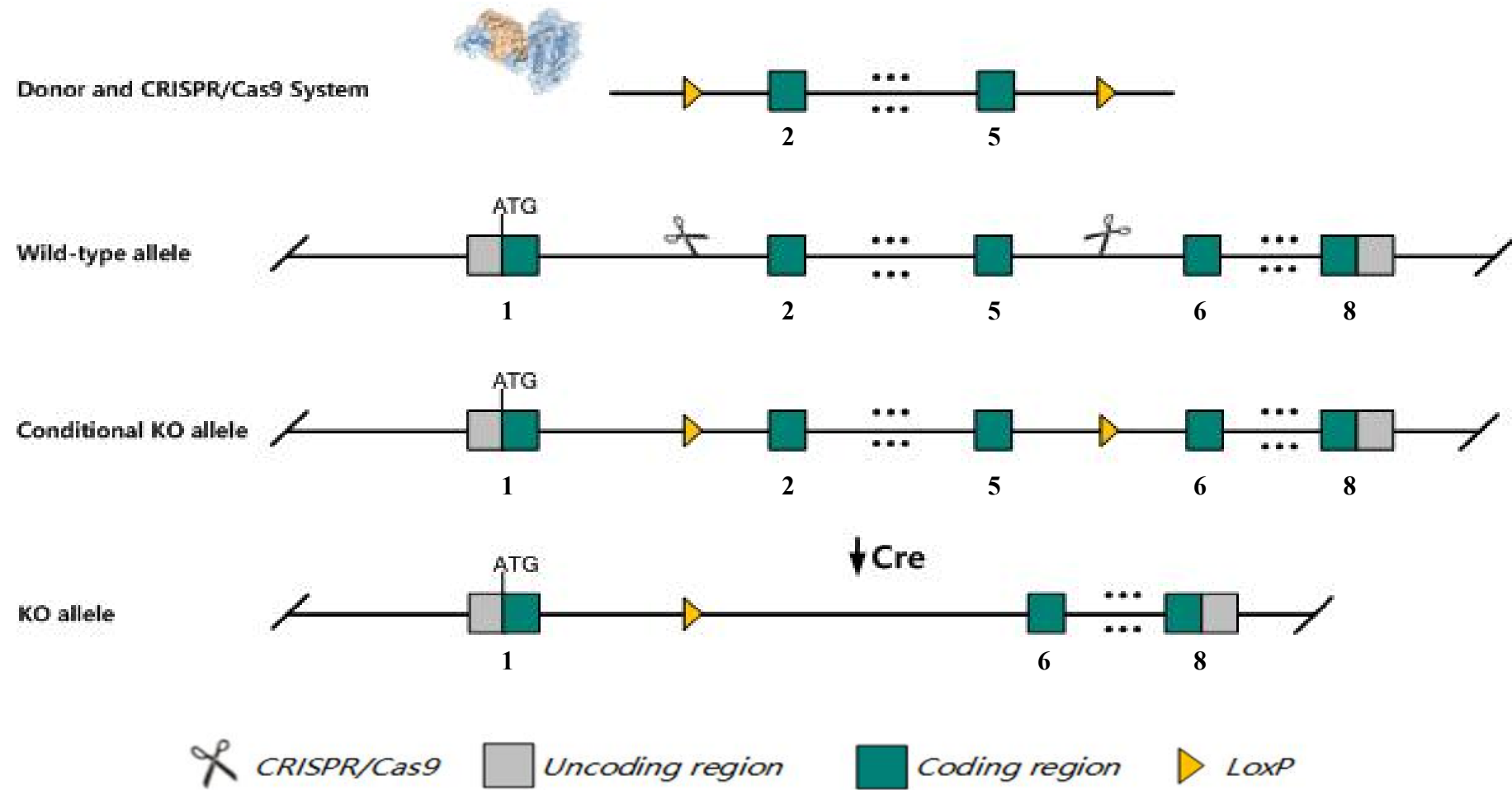
**Strain background**

**C57BL/6JGpt**

---

# Conditional Knockout strategy

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



The *Cytip* gene has 8 transcripts. According to the structure of *Cytip* gene, exon2-exon5 of *Cytip-201* (ENSMUST00000028175.6) transcript is recommended as the knockout region. The region contains 302bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Cytip* 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.

According to the existing MGI data, mice homozygous for a null allele display impaired trafficking and/or cell adhesion of immune system cells. mice homozygous for a reporter allele show normal immune cell development and function; however, mutant hematopoietic stem cells have impaired repopulating activity.

The *Cytip* gene is located on the Chr2. 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.

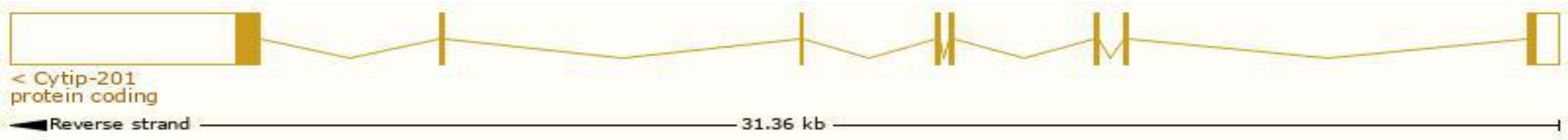


# Transcript information      Ensembl

The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cytip-201	<a href="#">ENSMUST00000028175.6</a>	6110	<a href="#">359aa</a>	Protein coding	<a href="#">CCDS16048</a>	<a href="#">Q91VY6</a>	TSL:1 GENCODE basic APPRIS P1
Cytip-205	<a href="#">ENSMUST00000148764.7</a>	941	No protein	Processed transcript	-	-	TSL:3
Cytip-203	<a href="#">ENSMUST00000144117.7</a>	856	No protein	Processed transcript	-	-	TSL:2
Cytip-204	<a href="#">ENSMUST00000146545.7</a>	856	No protein	Processed transcript	-	-	TSL:1
Cytip-207	<a href="#">ENSMUST00000151785.1</a>	686	No protein	Processed transcript	-	-	TSL:1
Cytip-202	<a href="#">ENSMUST00000131443.7</a>	641	No protein	Processed transcript	-	-	TSL:5
Cytip-206	<a href="#">ENSMUST00000151169.1</a>	387	No protein	Processed transcript	-	-	TSL:2
Cytip-208	<a href="#">ENSMUST00000153052.1</a>	378	No protein	Processed transcript	-	-	TSL:3

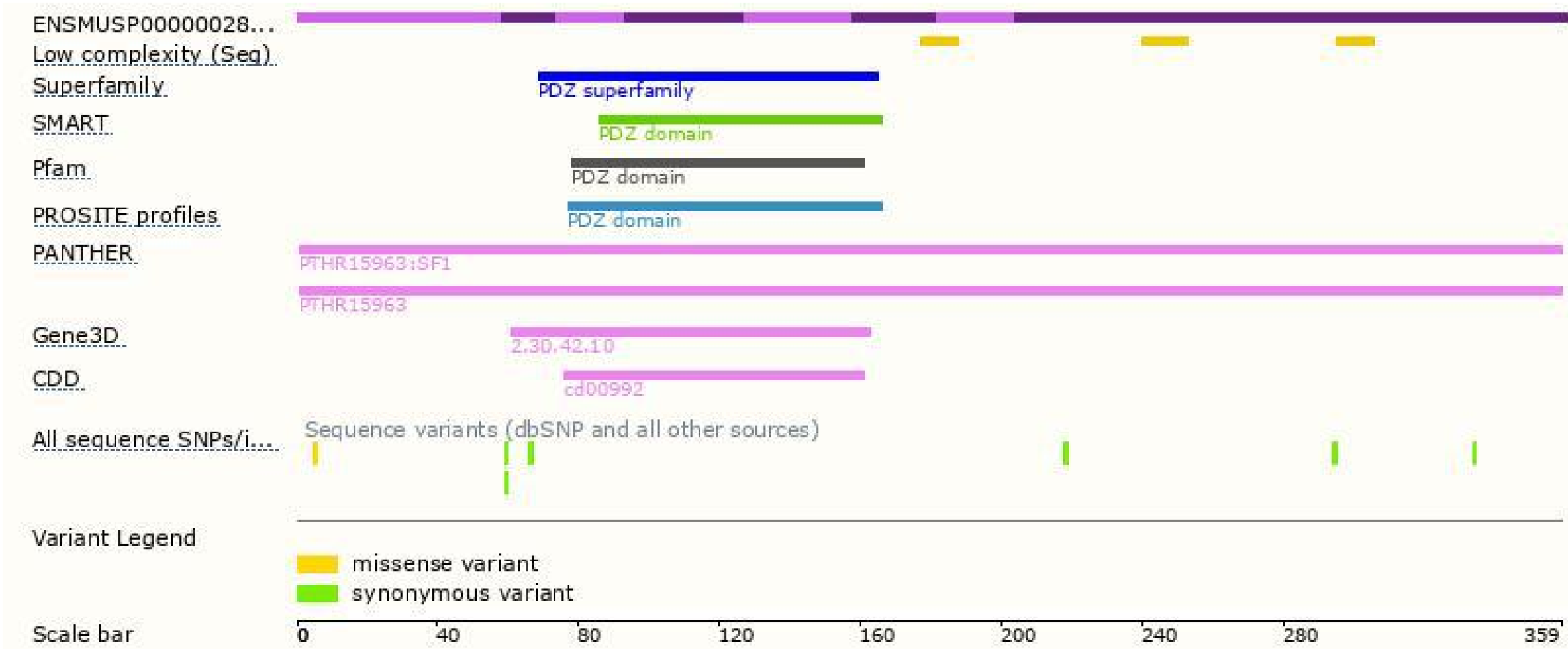
The strategy is based on the design of *Cytip-201* transcript,the transcription is shown below:



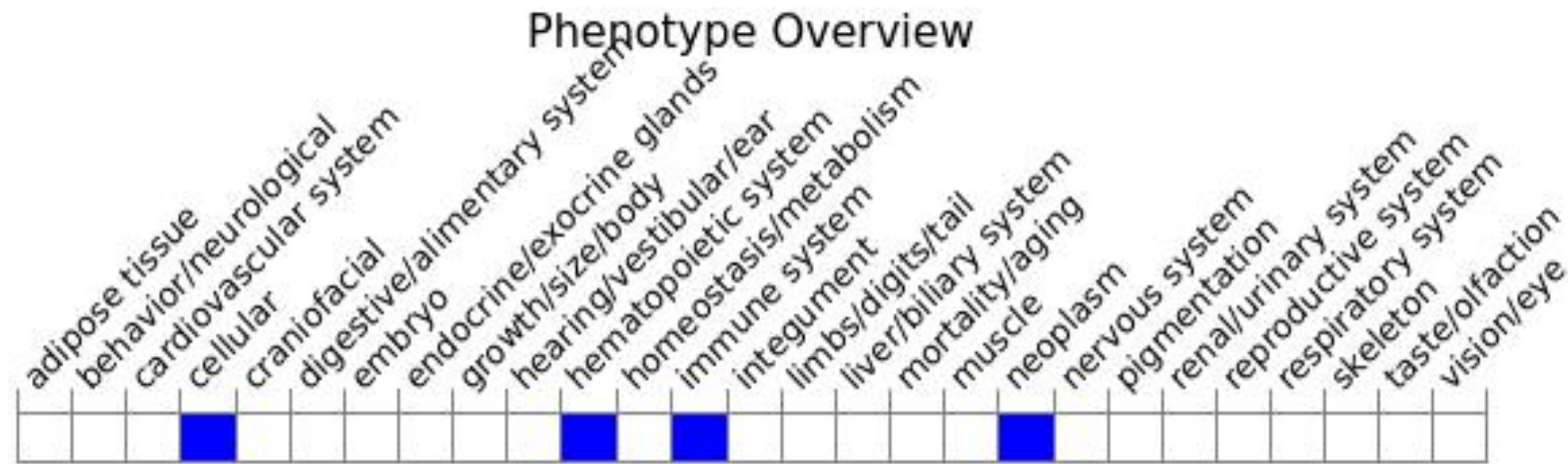
# Genomic location distribution



# 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 display impaired trafficking and/or cell adhesion of immune system cells. Mice homozygous for a reporter allele show normal immune cell development and function however, mutant hematopoietic stem cells have impaired repopulating activity.

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

