

Itgad Cas9-CKO Strategy

Designer:

JiaYu

Reviewer:

Xiaojing Li

Design Date:

2020-2-27

Project Overview

Project Name

Itgad

Project type

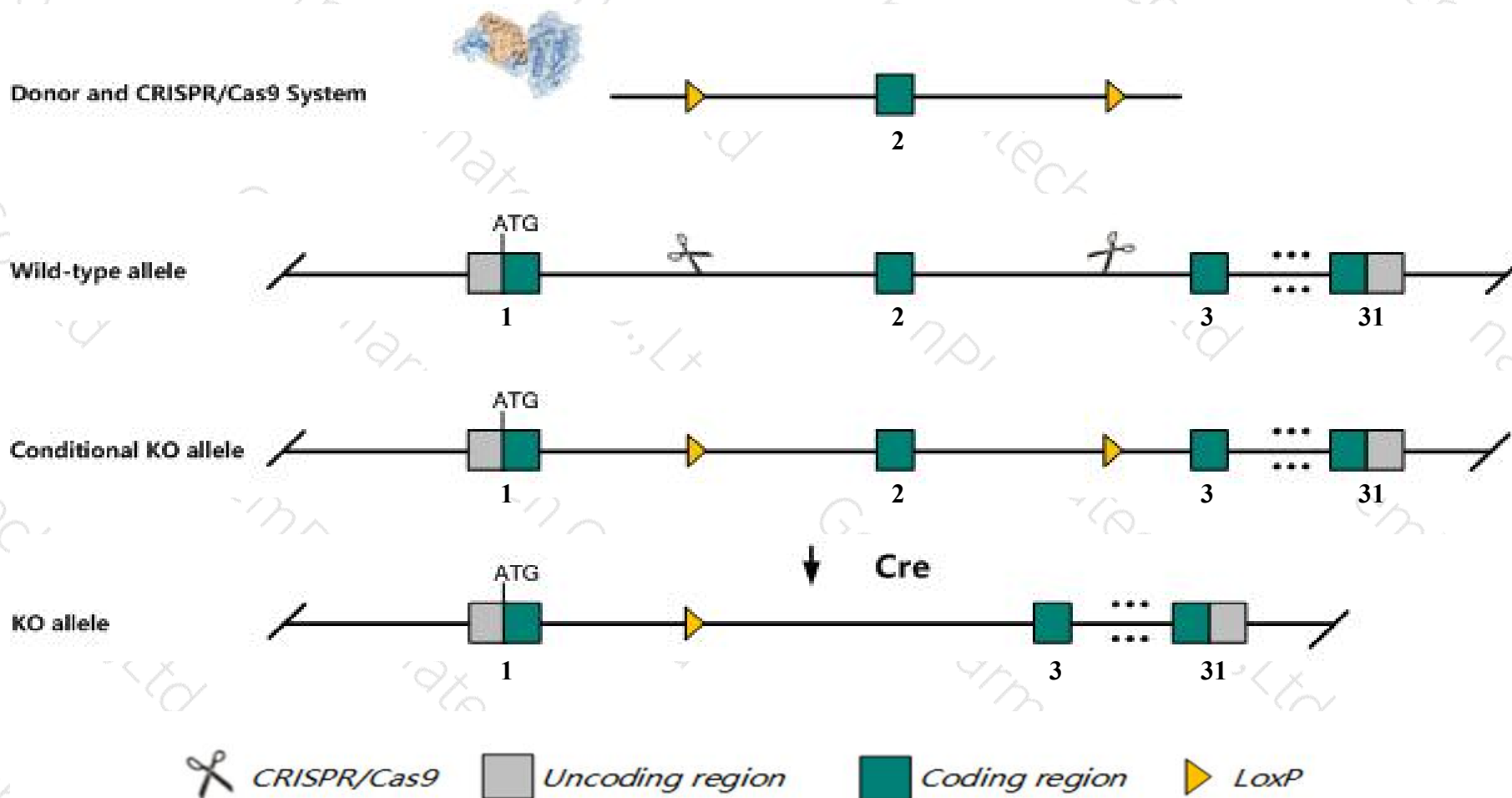
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- The *Itgad* gene has 5 transcripts. According to the structure of *Itgad* gene, exon2 of *Itgad-201* (ENSMUST00000033051.15) transcript is recommended as the knockout region. The region contains 106bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Itgad* 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, Homozygous null mice exhibit a reduced staphylococcal enterotoxin-induced T cell response.
- Transcript 203,205 CDS 5' incomplete the influences is unknown.
- The *Itgad* gene is located on the Chr7. 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)

Itgad integrin, alpha D [Mus musculus (house mouse)]

Gene ID: 381924, updated on 31-Jan-2019

Summary

Official Symbol	Itgad provided by MGI
Official Full Name	integrin, alpha D provided by MGI
Primary source	MGI:MGI:3578624
See related	Ensembl:ENSMUSG00000070369
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	Cd11d
Expression	Biased expression in spleen adult (RPKM 20.4), testis adult (RPKM 5.2) and 1 other tissue See more
Orthologs	human all

Transcript information (Ensembl)

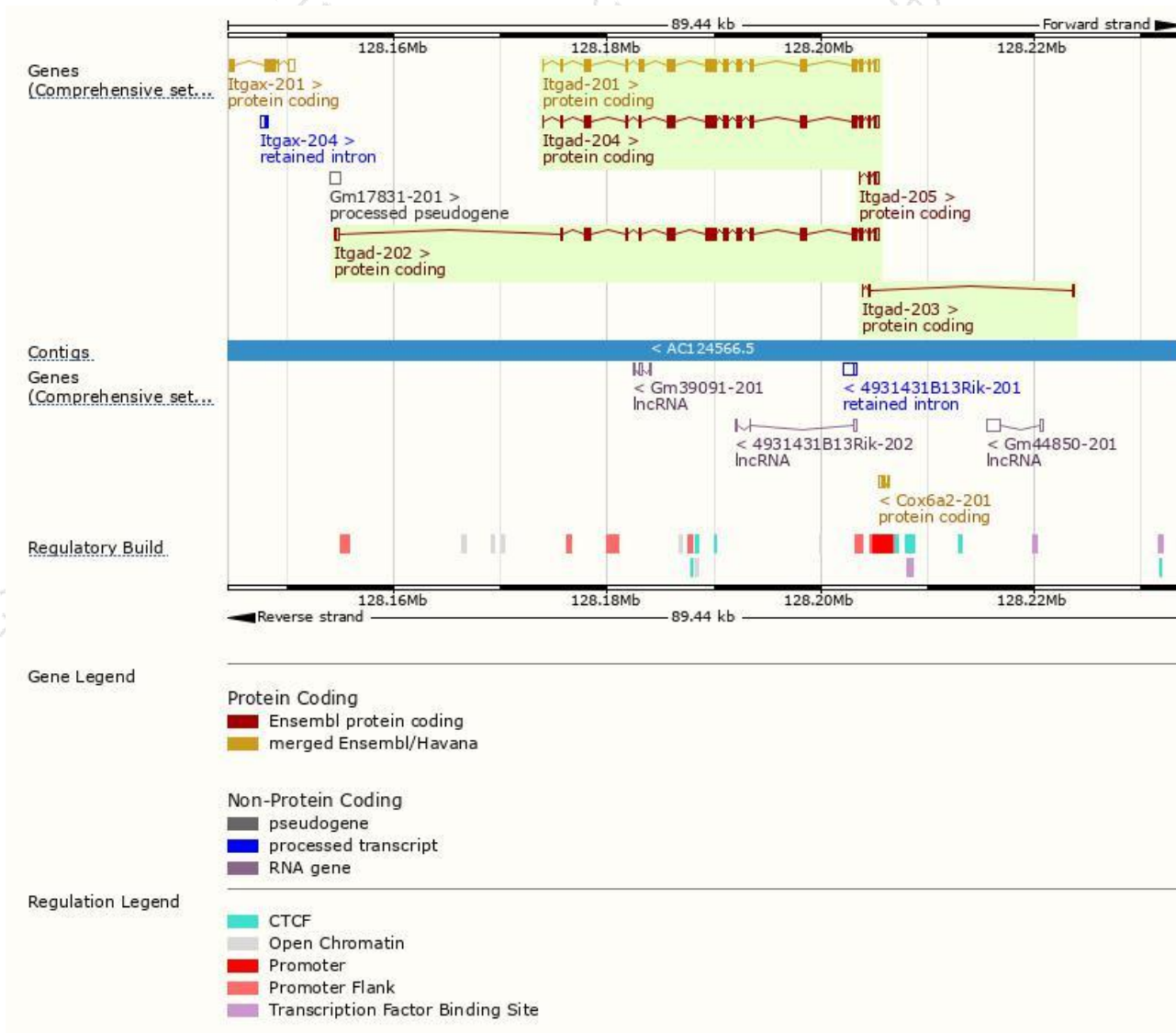
The gene has 5 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Itgad-201	ENSMUST00000033051.15	3965	1202aa	Protein coding	CCDS21890	E9PXZ7	TSL:1 GENCODE basic APPRIS P2
Itgad-202	ENSMUST00000106237.9	4105	1168aa	Protein coding	-	Q3V0T4	TSL:1 GENCODE basic APPRIS ALT2
Itgad-204	ENSMUST00000177111.7	3845	1166aa	Protein coding	-	H3BKX8	TSL:5 GENCODE basic APPRIS ALT2
Itgad-205	ENSMUST00000177383.7	653	107aa	Protein coding	-	H3BJS4	CDS 5' incomplete TSL:3
Itgad-203	ENSMUST00000176249.1	404	96aa	Protein coding	-	H3BKA0	CDS 5' incomplete TSL:2

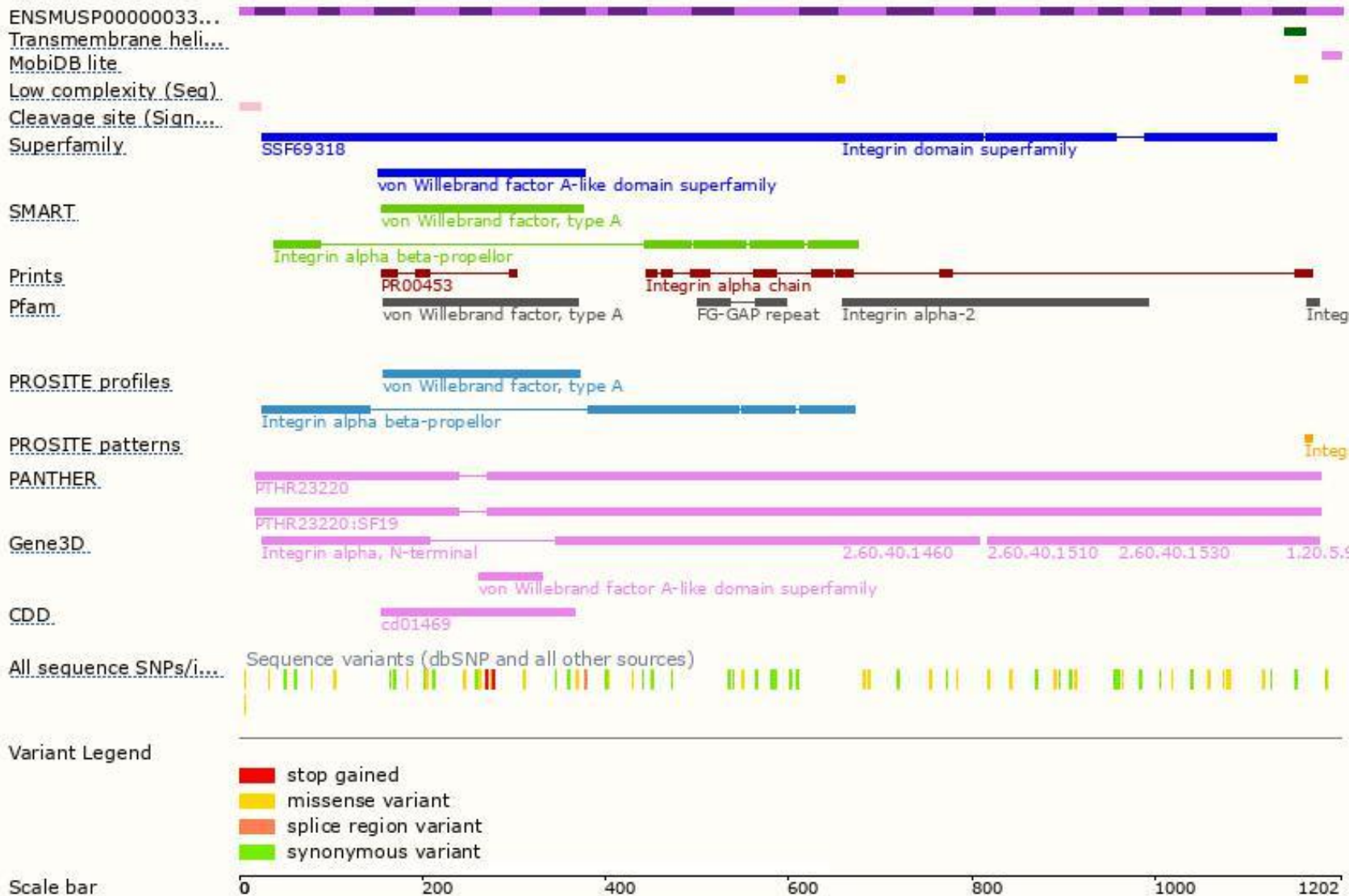
The strategy is based on the design of *Itgad-201* transcript, The transcription is shown below



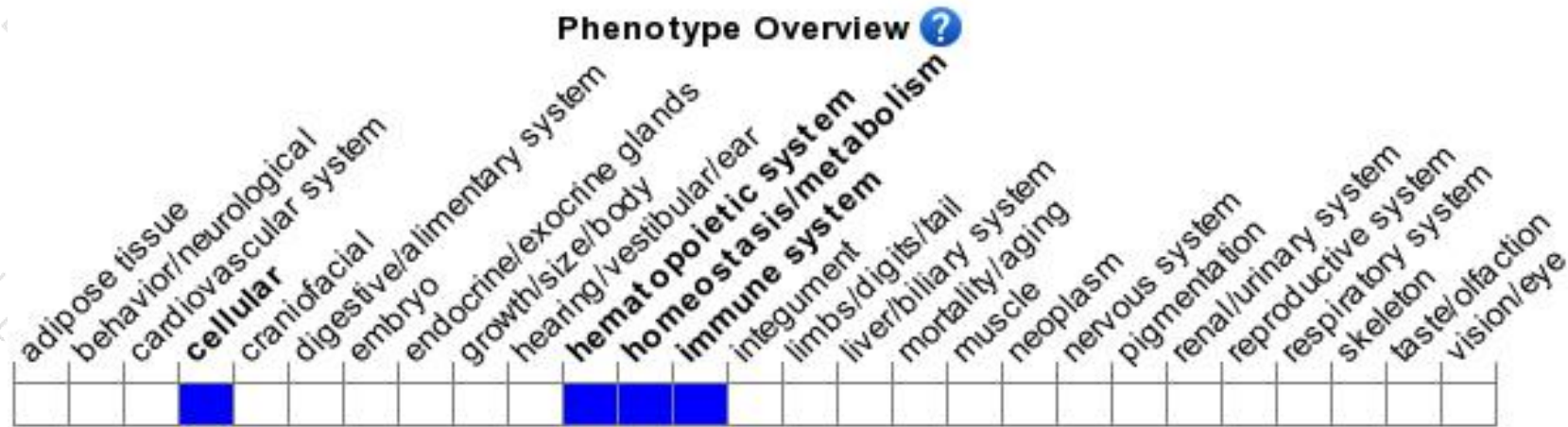
Genomic location distribution



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, Homozygous null mice exhibit a reduced staphylococcal enterotoxin-induced T cell response.

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

Tel: 400-9660890

