

# ***Itgb5* Cas9-CKO Strategy**

Designer: Xueting Zhang

# Project Overview

**Project Name**

*Itgb5*

**Project type**

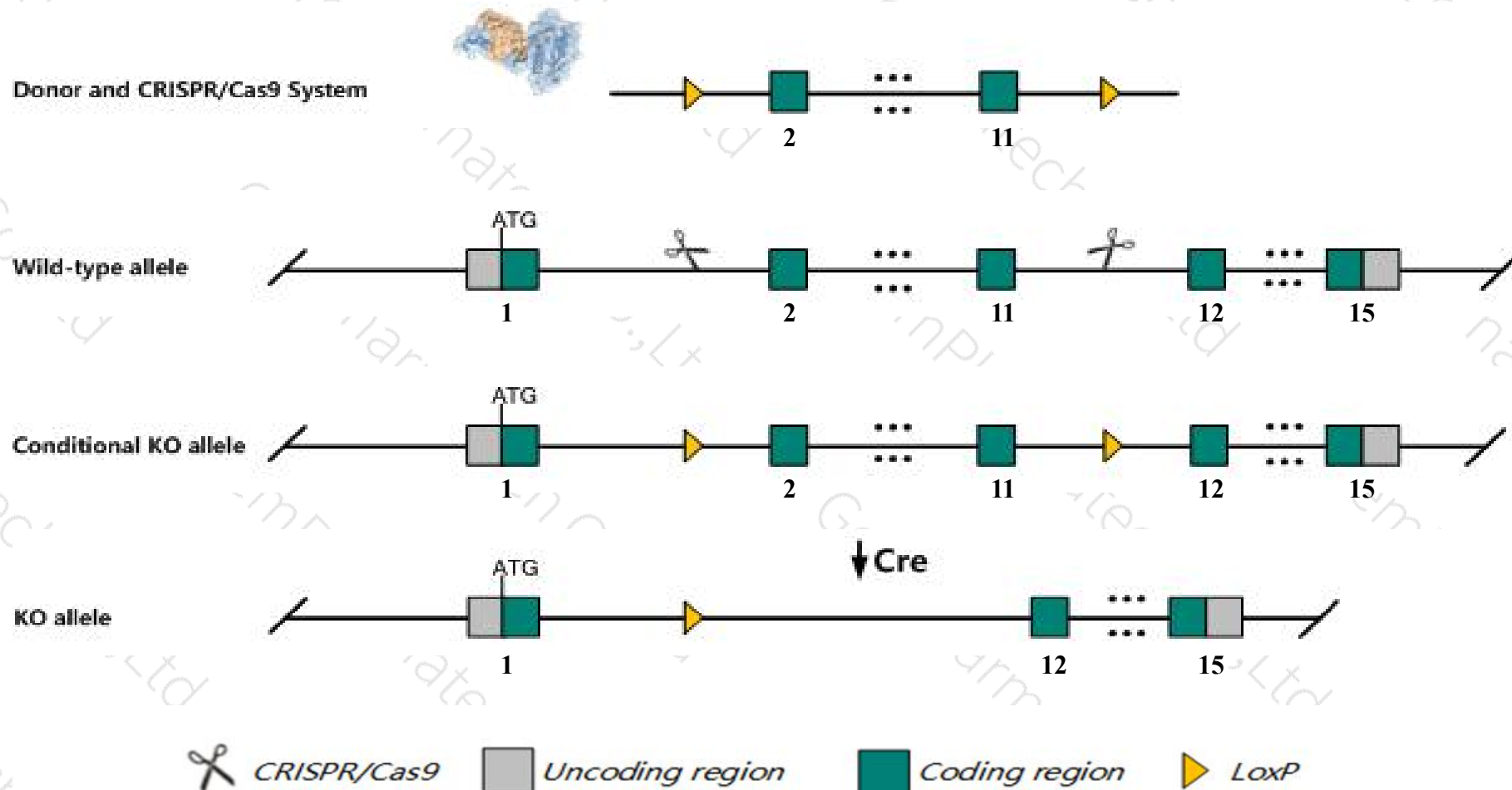
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Itgb5* gene. The schematic diagram is as follows:



- The *Itgb5* gene has 7 transcripts. According to the structure of *Itgb5* gene, exon2-exon11 of *Itgb5-201* (ENSMUST00000069345.5) transcript is recommended as the knockout region. The region contains 1846bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Itgb5* 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, Homozygotes for a targeted null mutation do not appear to differ from normal in respect to development, reproduction, adenovirus infection, or wound healing. Mutant keratinocytes do show reduced migration on, and adhesion to, vitronectin in vitro.
- Transcript *Itgb5*-206 may not be affected.
- The strategy will delete the start codon of transcrip *Itgb5*-206, which may form a new ATG and translate unknown proteins.
- The *Itgb5* gene is located on the Chr16. 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)

## Itgb5 integrin beta 5 [Mus musculus (house mouse)]

Gene ID: 16419, updated on 16-Feb-2019

### Summary



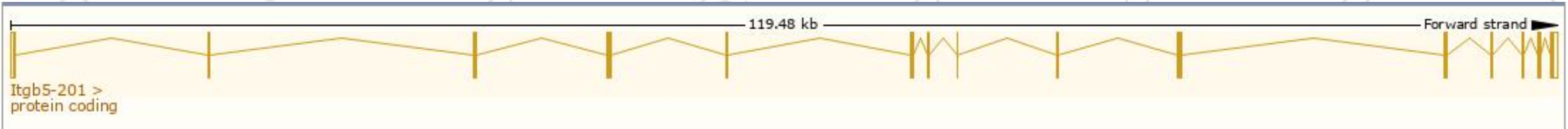
<b>Official Symbol</b>	Itgb5 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	integrin beta 5 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:96614</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000022817</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	VALIDATED
<b>Organism</b>	<a href="#">Mus musculus</a>
<b>Lineage</b>	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
<b>Also known as</b>	AA475909, AI874634, ESTM23, [b]-5, [b]5, [b]5A, [b]5B, beta-5, beta5
<b>Expression</b>	Ubiquitous expression in adrenal adult (RPKM 97.5), ovary adult (RPKM 81.3) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

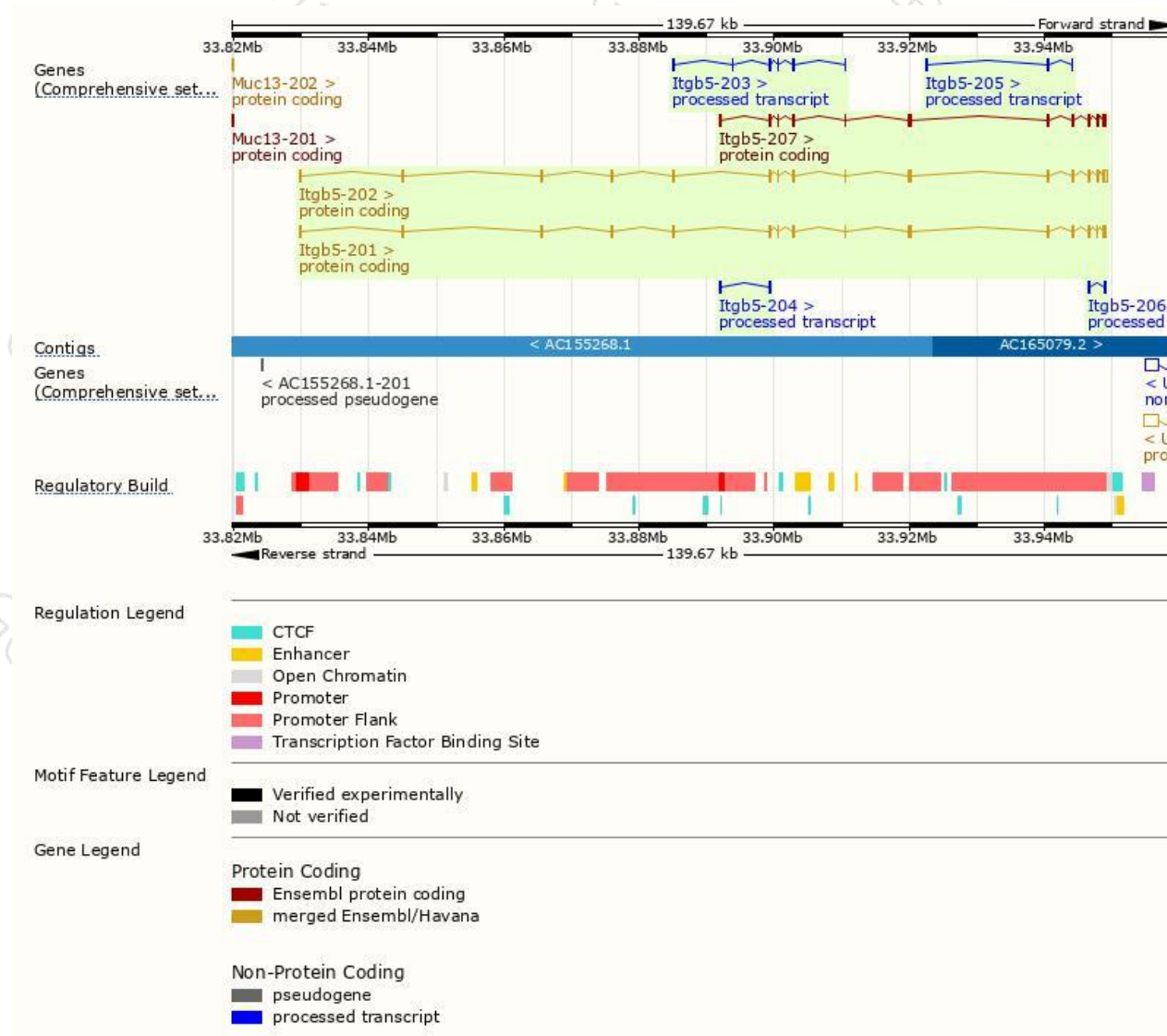
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Itgb5-202	<a href="#">ENSMUST00000115028.10</a>	3282	<a href="#">799aa</a>	Protein coding	<a href="#">CCDS49835</a>	<a href="#">Q6PE70</a>	TSL:1 GENCODE basic APPRIS P1
Itgb5-201	<a href="#">ENSMUST00000069345.5</a>	3056	<a href="#">816aa</a>	Protein coding	<a href="#">CCDS28135</a>	<a href="#">G5E8F8</a>	TSL:1 GENCODE basic
Itgb5-207	<a href="#">ENSMUST00000232262.1</a>	2130	<a href="#">486aa</a>	Protein coding	-	<a href="#">A0A338P795</a>	GENCODE basic
Itgb5-203	<a href="#">ENSMUST00000134262.7</a>	631	No protein	Processed transcript	-	-	TSL:2
Itgb5-205	<a href="#">ENSMUST00000151930.1</a>	364	No protein	Processed transcript	-	-	TSL:3
Itgb5-206	<a href="#">ENSMUST00000231409.1</a>	297	No protein	Processed transcript	-	-	
Itgb5-204	<a href="#">ENSMUST00000148462.1</a>	265	No protein	Processed transcript	-	-	TSL:5

The strategy is based on the design of *Itgb5-201* transcript,The transcription is shown below



# Genomic location distribution

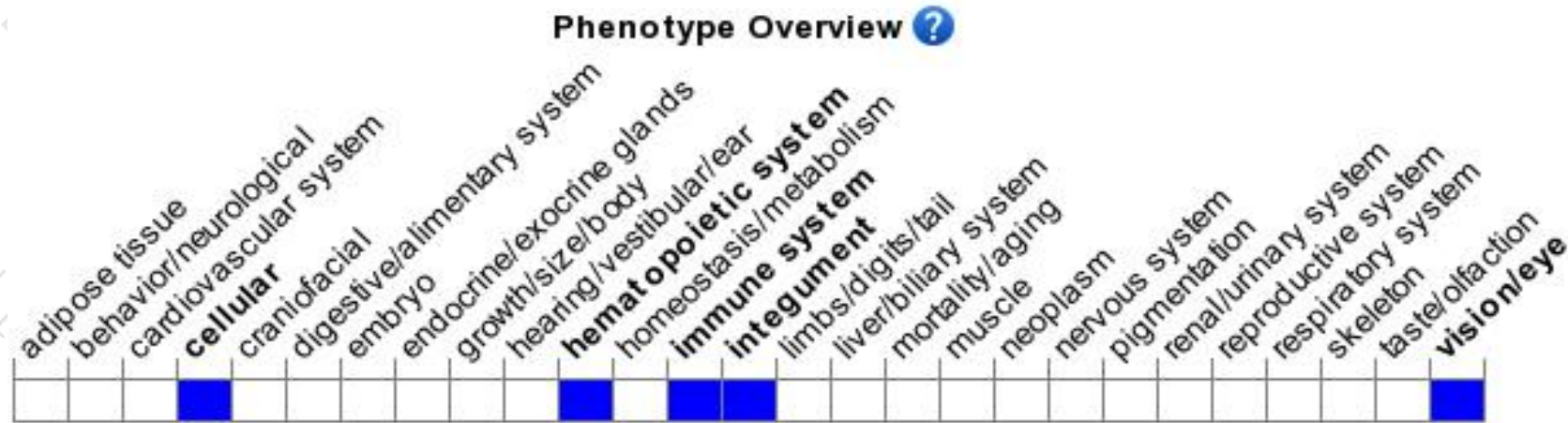




# Protein domain



# 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 do not appear to differ from normal in respect to development, reproduction, adenovirus infection, or wound healing. Mutant keratinocytes do show reduced migration on, and adhesion to, vitronectin in vitro.

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

Tel: 400-9660890

