

Il6 Cas9-KO Strategy

Designer: Jing Jin

Reviewer: Yang Zeng

Design Date: 2018-6-21

Project Overview



Project Name 116

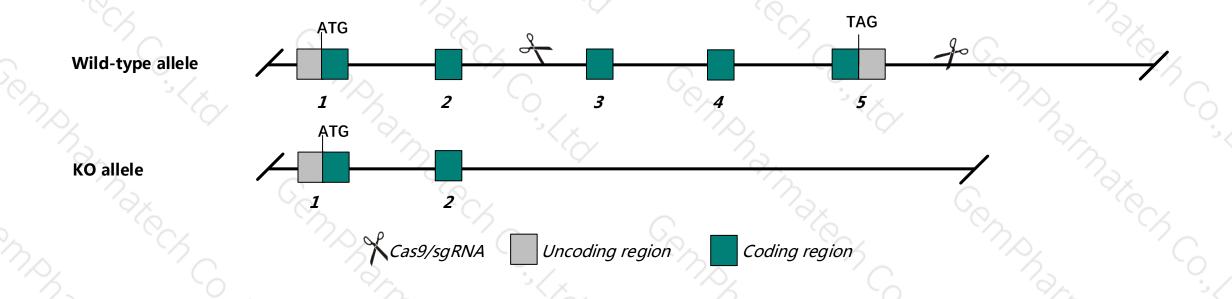
Project type Cas9-KO

Strain background C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Il6* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Il6* gene has 4 transcripts. According to the structure of *Il6* gene, exon3-exon5 of *Il6-201* (ENSMUST00000026845.11) transcript is recommended as the knockout region. The region contains 432bp coding sequence and 3'UTR region.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Il6* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice



- > According to the existing MGI data, Homozygous null mutants show impaired immune response to pathogens, decreased T cell numbers and resistance to plasma cell neoplasia. They are defective in wound healing and liver regeneration and show increased emotionality and high bone turnover rate.
- The *Il6* is close to 5'UTR region of the *A230098N10Rik* gene. Knockout the region may affect the function of the *A230098N10Rik* gene.
- ➤ The *Il6* gene is located on the Chr5. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



ll6 interleukin 6[*Mus musculus* (house mouse)]

Gene ID: 16193, updated on 13-Aug-2019

Summary

☆ ?

Official Symbol II6 provided by MGI

Official Full Name interleukin 6 provided by MGI

Primary source MGI:MGI:96559

See related Ensembl:ENSMUSG00000025746

Gene type protein coding
RefSeq status REVIEWED
Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as II-6

Summary This gene encodes a member of the interleukin family of cytokines that have important functions in immune response,

hematopoiesis, inflammation and the acute phase response. The ectopic overexpression of the encoded protein in mice results

in excessive plasma cells in circulation, leading to death. Mice lacking the encoded protein exhibit abnormalities in hepatic acute phase response, some immune mechanisms, bone resorption in response to estrogen, liver regeneration and wound

healing. Alternative splicing results in multiple transcript variants encoding different isoforms. [provided by RefSeq, Sep 2015]

Expression Low expression observed in reference dataset <u>See more</u>

Orthologs <u>human</u> all

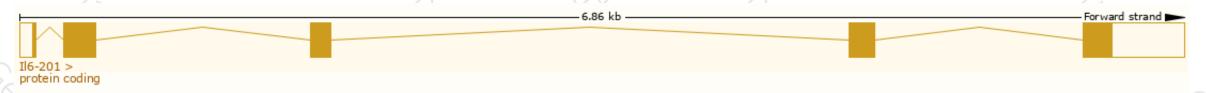
Transcript information (Ensembl)



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

Show/hide columns Filter								
Name A	Transcript ID 🔷	bp 🏺	Protein 🌲	Translation ID 🔷	Biotype	CCDS 🍦	UniProt 🛊	Flags 🔷
116-201	ENSMUST00000026845.11	1141	<u>211aa</u>	ENSMUSP00000026845.7	Protein coding	CCDS19153 ₽	<u>A2RTD1</u> & <u>P08505</u> &	TSL:1 GENCODE basic APPRIS P2
116-202	ENSMUST00000195978.4	651	<u>165aa</u>	ENSMUSP00000143544.1	Protein coding	CCDS84856 ₺	A0A0G2JGF4 &	TSL:1 GENCODE basic
116-203	ENSMUST00000199183.4	2343	<u>208aa</u>	ENSMUSP00000143293.1	Protein coding	-	A0A0G2JFT1 &	TSL:2 GENCODE basic
116-204	ENSMUST00000199765.1	963	<u>194aa</u>	ENSMUSP00000143157.1	Protein coding	-	A0A0G2JFG1₺	TSL:2 GENCODE basic APPRIS ALT2

The strategy is based on the design of *Il6-201* transcript, The transcription is shown below



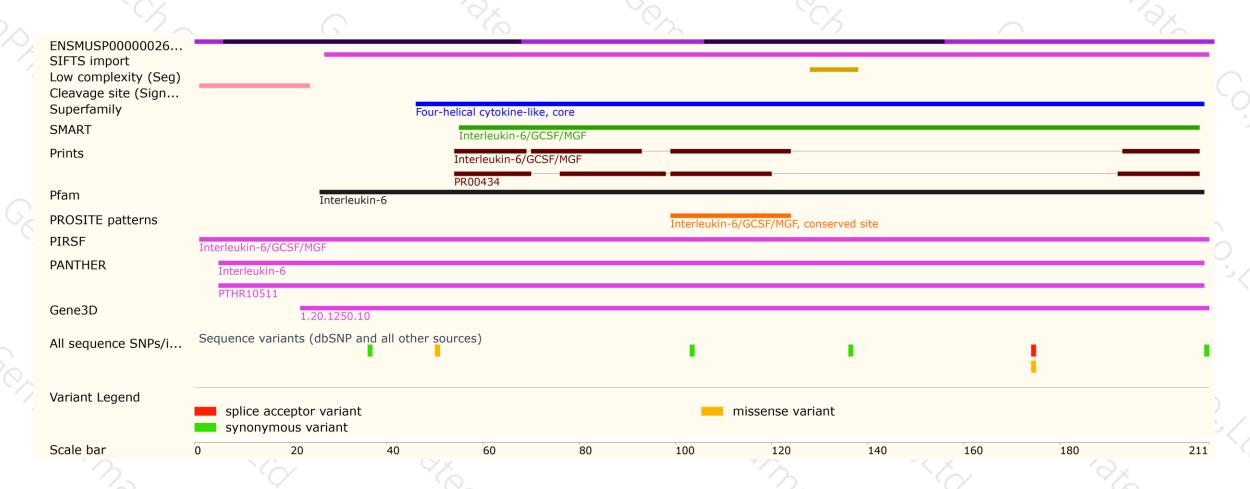
Genomic location distribution





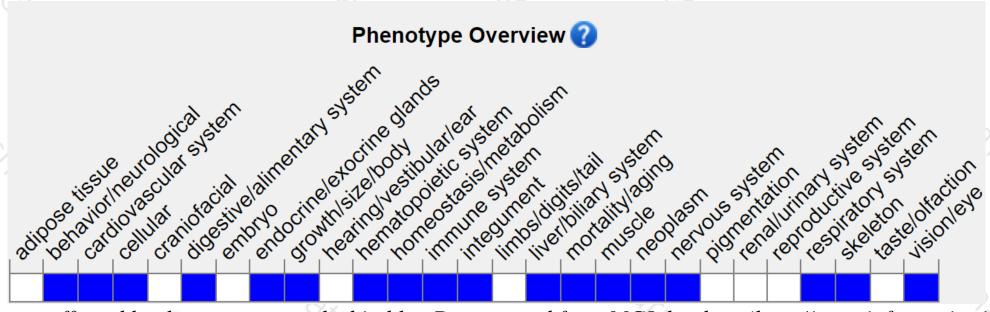
Protein domain





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 mutants show impaired immune response to pathogens, decreased T cell numbers and resistance to plasma cell neoplasia. They are defective in wound healing and liver regeneration and show increased emotionality and high bone turnover rate.



If you have any questions, you are welcome to inquire. Tel: 025-5864 1534





