

Igfbp2 Cas9-CKO Strategy

Designer:

Daohua Xu

Reviewer:

Huimin Su

Design Date:

2019-9-28

Project Overview



Project Name

Igfbp2

Project type

Cas9-CKO

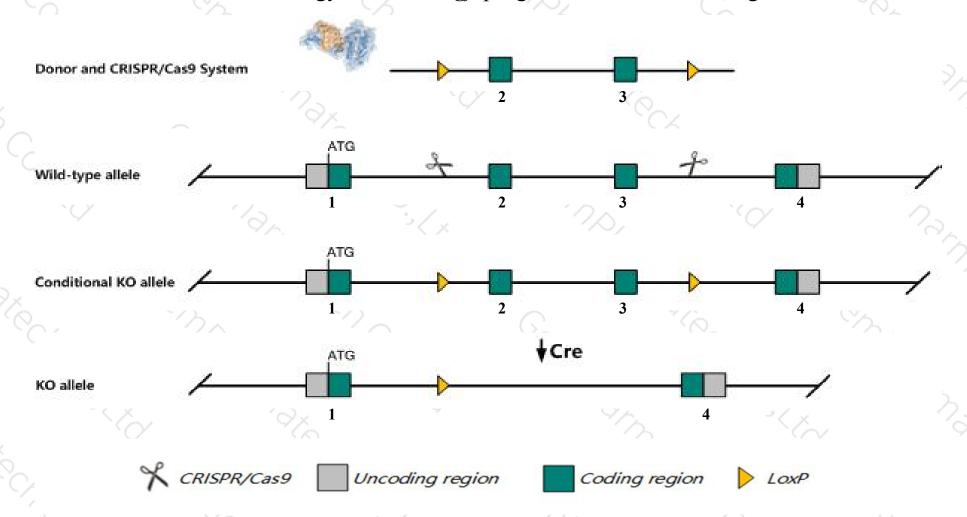
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Igfbp2* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Igfbp2* gene has 3 transcripts. According to the structure of *Igfbp2* gene, exon2-exon3 of *Igfbp2-201*(ENSMUST00000047328.10) transcript is recommended as the knockout region. The region contains 368bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Igfbp2* 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, Homozygous mutation of this gene results in reduced spleen, heart and kidney size and increased liver weight. Homozygotes for another allele exhibit a normal phenotype.
- The *Igfbp2* gene is located on the Chr1. 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)



Igfbp2 insulin-like growth factor binding protein 2 [Mus musculus (house mouse)]

Gene ID: 16008, updated on 2-Apr-2019

Summary

↑ ?

Official Symbol Igfbp2 provided by MGI

Official Full Name insulin-like growth factor binding protein 2 provided by MGI

Primary source MGI:MGI:96437

See related Ensembl: ENSMUSG00000039323

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 Al255832, IBP-2, Igfbp-2, mIGFBP-2

Summary The protein encoded by this gene is one of several similar proteins that bind insulin-like growth factors I and II (lgf-I and lgf-II). The encoded

protein can be secreted into the bloodstream, where it binds Igf-I and Igf-II with high affinity, or it can remain intracellular, interacting with many different ligands. Two transcript variants, one encoding a secreted isoform and the other encoding a nonsecreted isoform, have been

found for this gene. [provided by RefSeq, Sep 2015]

Expression Biased expression in bladder adult (RPKM 311.4), liver E18 (RPKM 285.8) and 12 other tissuesSee more

Orthologs <u>human</u> all

Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
lgfbp2-201	ENSMUST00000047328.10	1307	305aa	Protein coding	CCDS15036	P47877	TSL:1 GENCODE basic APPRIS P1
lgfbp2-202	ENSMUST00000120564.1	991	<u>158aa</u>	Protein coding	CCDS78613	D3YU40	TSL:3 GENCODE basic
lgfbp2-203	ENSMUST00000155703.1	906	No protein	Retained intron	20	-	TSL:2

The strategy is based on the design of *Igfbp2-201* transcript, The transcription is shown below

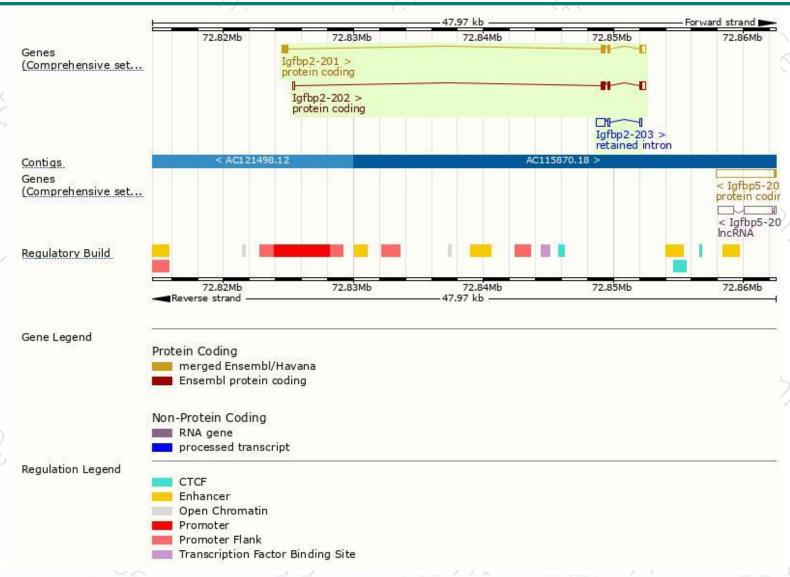




27.97 kb

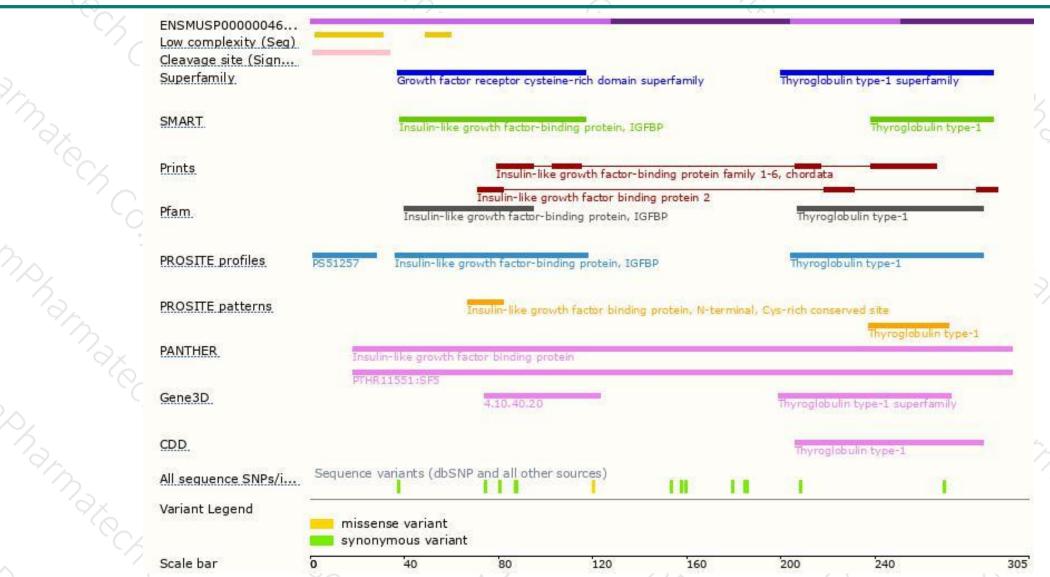
Genomic location distribution





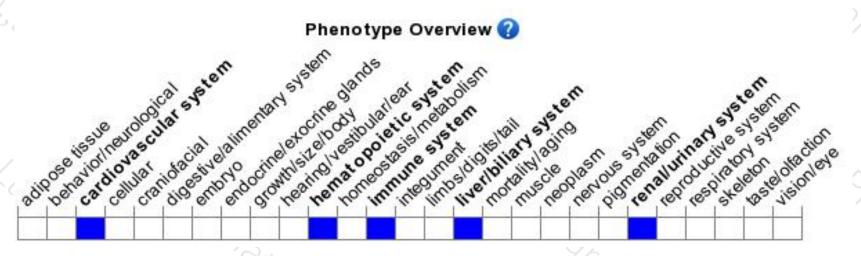
Protein domain





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 mutation of this gene results in reduced spleen, heart and kidney size and increased liver weight. Homozygotes for another allele exhibit a normal phenotype.



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





