

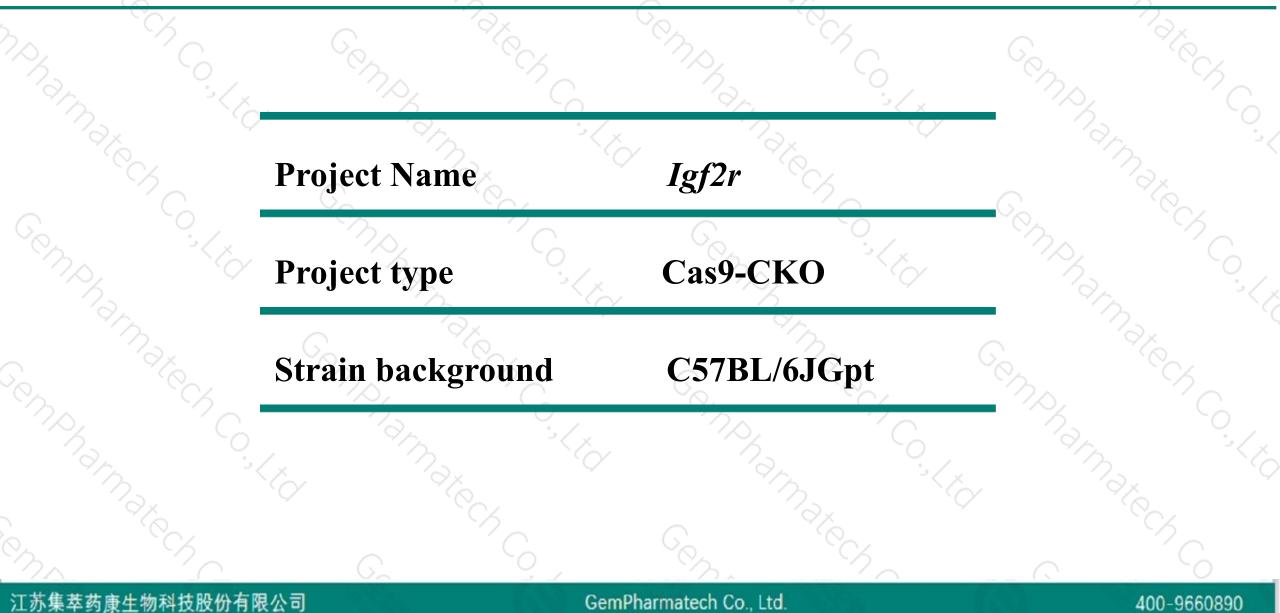
Igf2r Cas9-CKO Strategy

Designer: Xueting Zhang Design Date: 2019-7-25

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Project Overview

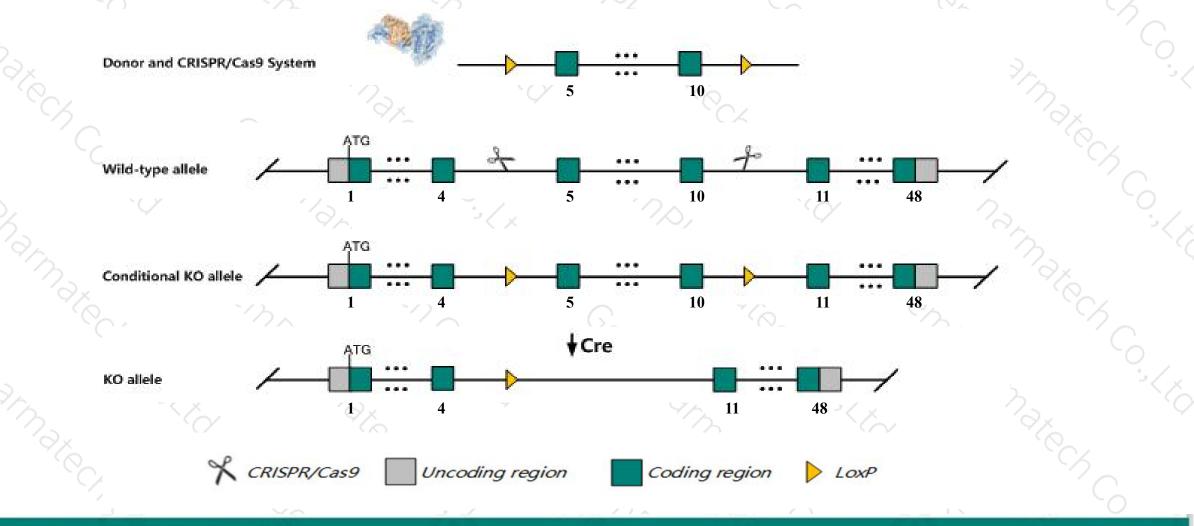




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Igf2r* gene. The schematic diagram is as follows:



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The *Igf2r* gene has 3 transcripts. According to the structure of *Igf2r* gene, exon5-exon10 of *Igf2r-201* (ENSMUST0000024599.13) transcript is recommended as the knockout region. The region contains 802bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Igf2r* 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, Mutants inheriting maternally a targeted disruption of this gene exhibit elevated serum and tissue IGF-II levels, overgrowth, organomegaly, kinky tail, polydactyly, heart defects, edema, dyspnea, imperforate vagina, reduced fertility and perinatal death. Survival is influenced by genetic background.
- ➤ Transcript *Igf2*r-202&203 may not be affected.
- > The N-terminal of Igf2r gene will remain 166aa, it may remain the partial function of Igf2r gene.
- The floxed region is near to the N-terminal of *Aim* gene, this strategy may influence the regulatory function of the N-terminal of *Aim* gene.
- The Igf2r gene is located on the Chr17. 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

Gene information (NCBI)



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Igf2r insulin-like growth factor 2 receptor [Mus musculus (house mouse)]

Gene ID: 16004, updated on 7-Apr-2019

Summary

Official Symbol	Igf2r provided by MGI
Official Full Name	insulin-like growth factor 2 receptor provided by MGI
Primary source	MGI:MGI:96435
See related	Ensembl:ENSMUSG0000023830
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	Al661837, CD222, CI-MPR, M6P/IGF2R, Mpr300
Expression	Ubiquitous expression in limb E14.5 (RPKM 34.0), placenta adult (RPKM 24.7) and 28 other tissuesSee more
Orthologs	human all

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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
lgf2r-201	ENSMUST00000024599.13	8887	<u>2483aa</u>	Protein coding	CCDS37436	<u>Q07113</u>	TSL:1 GENCODE basic APPRIS P1
lgf2r-203	ENSMUST00000161738.1	803	No protein	Processed transcript	-8		TSL:2
lgf2r-202	ENSMUST00000159127.1	245	No protein	Processed transcript	-	-	TSL:1

The strategy is based on the design of *Igf2r-201* transcript, The transcription is shown below

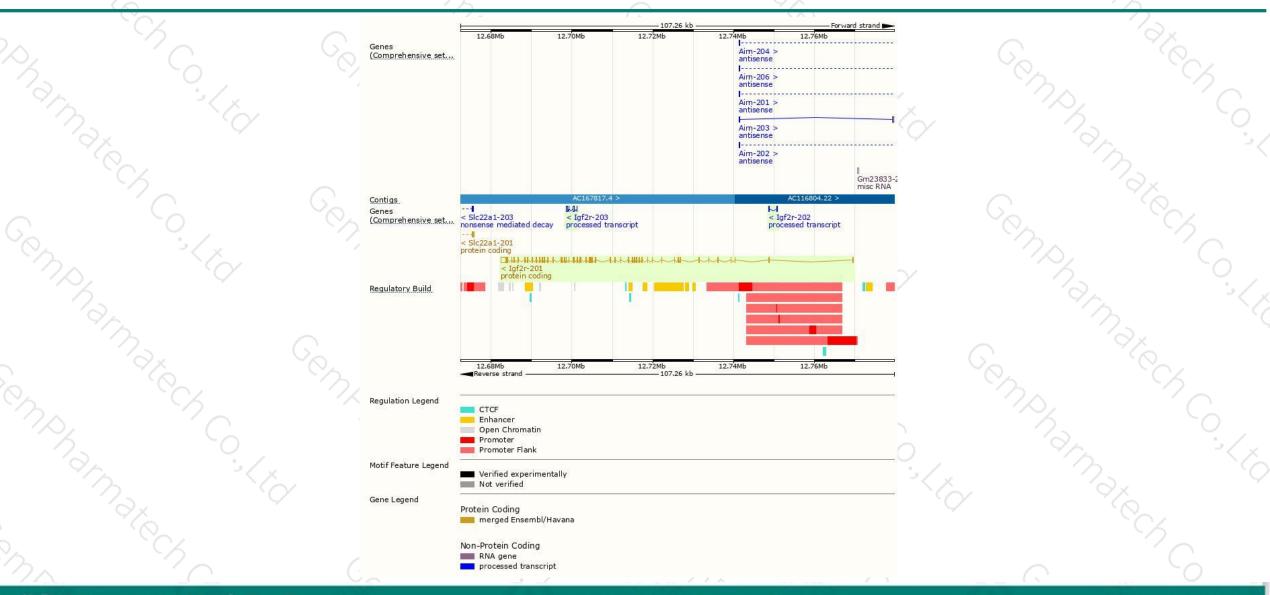
< Igf2r-201 protein coding

Reverse strand

- 87.26 kb

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Genomic location distribution



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Protein domain



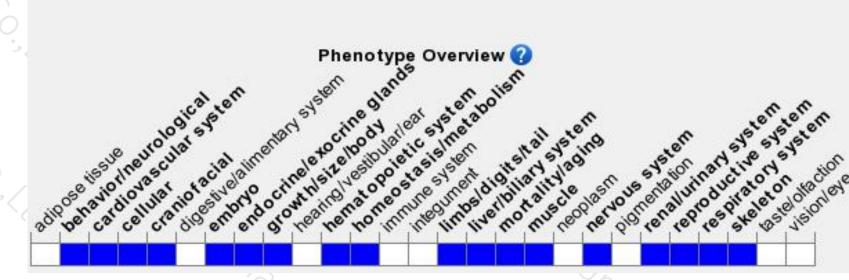


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Mouse phenotype description(MGI)





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, Mutants inheriting maternally a targeted disruption of this gene exhibit elevated serum and tissue IGF-II levels, overgrowth, organomegaly, kinky tail, polydactyly, heart defects, edema, dyspnea, imperforate vagina, reduced fertility and perinatal death. Survival is influenced by genetic background.

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



