

Cxcl12 Cas9-CKO Strategy

Designer:

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Design Date:

2019-10-18

Project Overview



Project Name

Cxcl12

Project type

Cas9-CKO

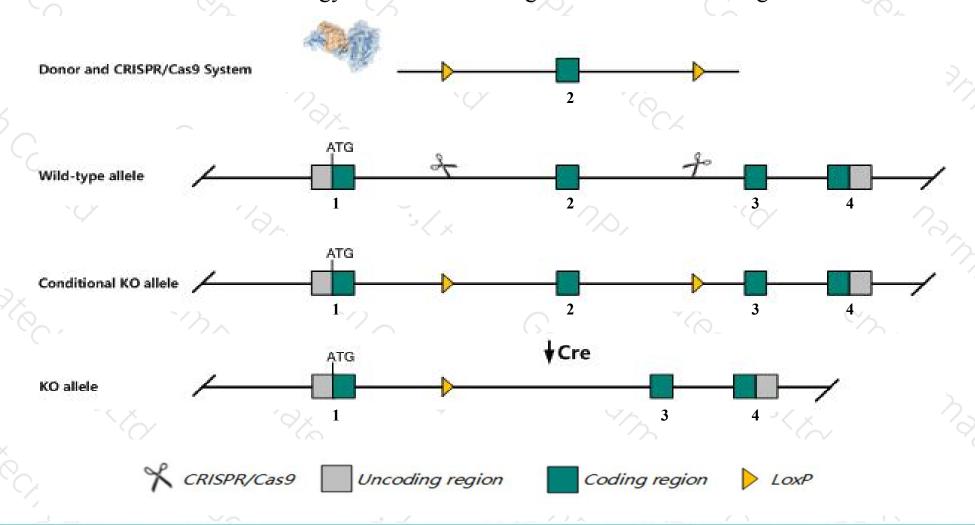
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cxcl12* gene. The schematic diagram is as follows:



Technical routes



- The *Cxcl12* gene has 4 transcripts. According to the structure of *Cxcl12* gene, exon2 of *Cxcl12-201*(ENSMUST00000073043.4) transcript is recommended as the knockout region. The region contains 118bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Cxcl12* 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 null mice display late embryonic lethality, impaired myelopoiesis, abnormal cerebellum development, abnormal germ cell migration, abnormal angiogenesis around the stomach, and ventricular septal defects.
- > The Cxcl12 gene is located on the Chr6. 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)



Cxcl12 chemokine (C-X-C motif) ligand 12 [Mus musculus (house mouse)]

Gene ID: 20315, updated on 9-Apr-2019

Summary

☆ ?

Official Symbol Cxcl12 provided by MGI

Official Full Name chemokine (C-X-C motif) ligand 12 provided by MGI

Primary source MGI:MGI:103556

See related Ensembl:ENSMUSG00000061353

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 Pbsf, Scyb12, Sdf1, Tlsf, Tpar1

Summary This gene encodes a member of the alpha chemokine protein family. The encoded protein is secreted and functions as the ligand for the G-

protein coupled receptor, chemokine (C-X-C motif) receptor 4. The encoded protein plays a role in many diverse cellular functions, including embryogenesis, immune surveillance, inflammation response, tissue homeostasis, and tumor growth and metastasis. Alternative splicing

results in multiple transcript variants. [provided by RefSeq, May 2013]

Expression Ubiquitous expression in spleen adult (RPKM 77.0), liver adult (RPKM 50.9) and 27 other tissues See more

Orthologs <u>human</u> all

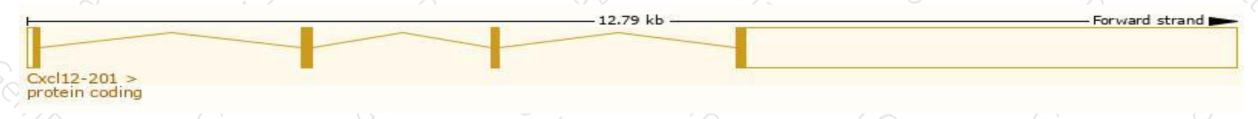
Transcript information (Ensembl)



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

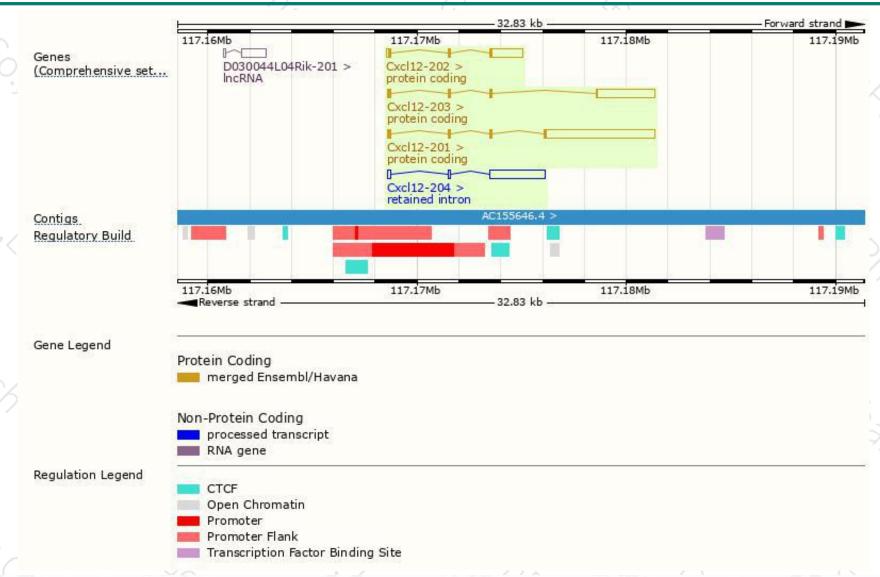
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cxcl12-201	ENSMUST00000073043.4	5627	<u>119aa</u>	Protein coding	CCDS20463	H7BX38	TSL:1 GENCODE basic
Cxcl12-203	ENSMUST00000112871.7	3132	<u>93aa</u>	Protein coding	CCDS39605	P40224	TSL:1 GENCODE basic APPRIS P4
Cxcl12-202	ENSMUST00000112866.7	1878	89aa	Protein coding	CCDS39606	P40224	TSL:1 GENCODE basic APPRIS ALT1
Cxcl12-204	ENSMUST00000134244.1	2894	No protein	Retained intron	9	12	TSL:1

The strategy is based on the design of Cxcl12-201 transcript, The transcription is shown below



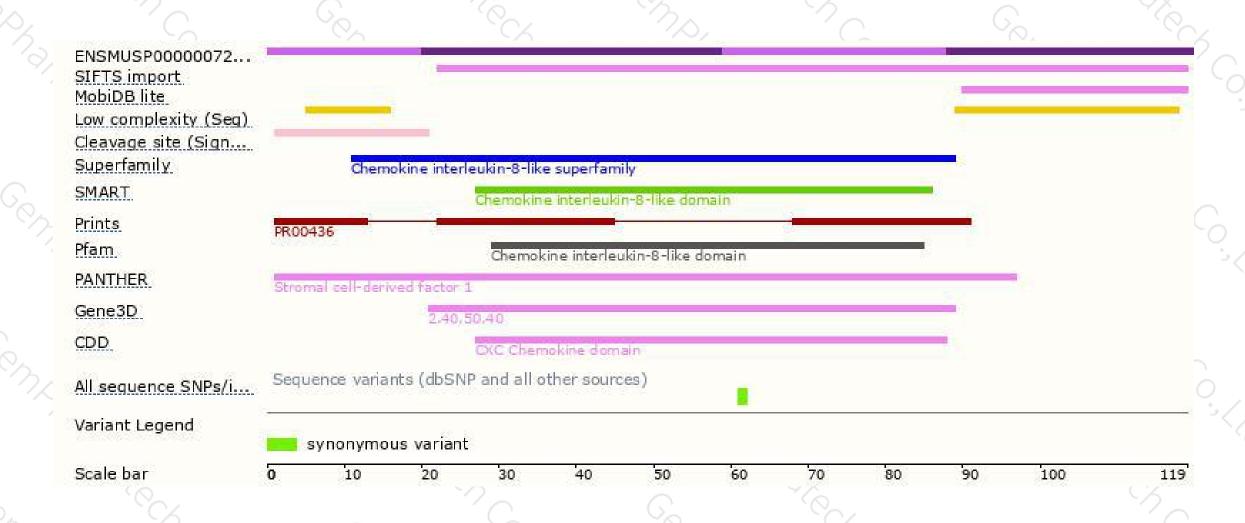
Genomic location distribution





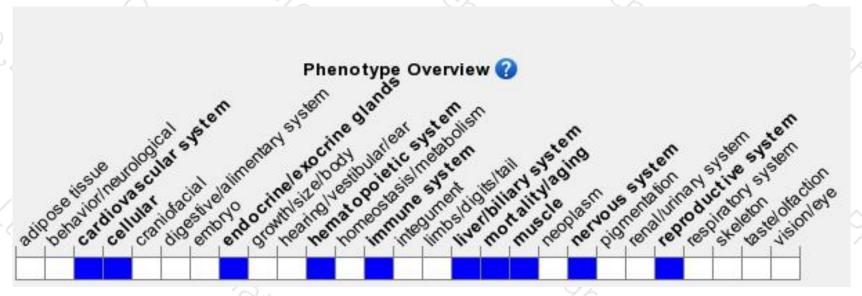
Protein domain





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 display late embryonic lethality, impaired myelopoiesis, abnormal cerebellum development, abnormal germ cell migration, abnormal angiogenesis around the stomach, and ventricula septal defects.



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





