

Bmp8b Cas9-CKO Strategy

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Project Overview

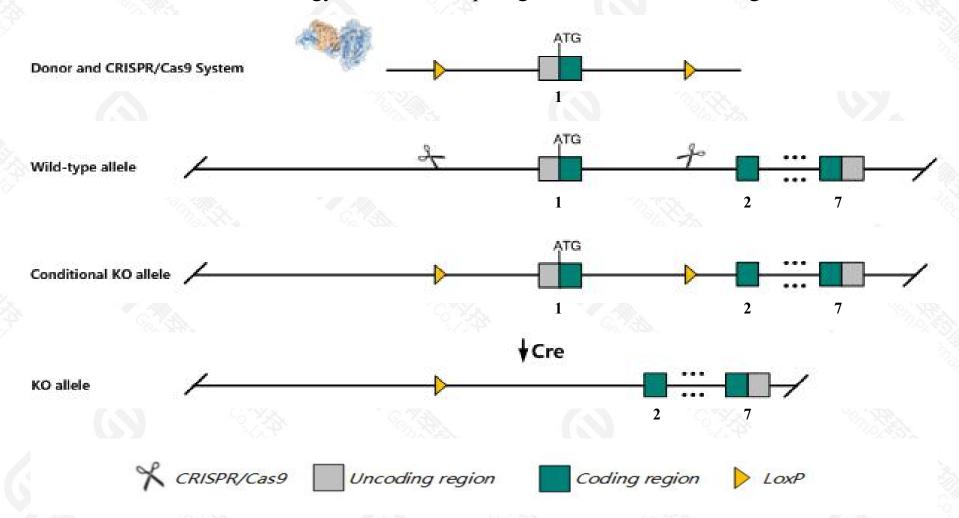


Project Name	Bmp8b			
Project type	Cas9-CKO			
Strain background	C57BL/6JGpt			

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Bmp8b* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Bmp8b* gene has 2 transcripts. According to the structure of *Bmp8b* gene, exon1 of *Bmp8b*201(ENSMUST00000002457.2) transcript is recommended as the knockout region. The region contains start codon
 ATG.Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Bmp8b* 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, incidence of lethality among homozygous null mutants is variable depending on genetic background and due to allantoic and embryonic abnormalities. Heterozygous and surviving homozygous males exhibit varying degrees of germ cell deficiency and infertility, also background dependent.
- > The *Bmp8b* gene is located on the Chr4. 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)



Bmp8b bone morphogenetic protein 8b [Mus musculus (house mouse)]

Gene ID: 12164, updated on 13-Mar-2020

Summary

Official Symbol Bmp8b provided by MGI

Official Full Name bone morphogenetic protein 8b provided by MGI

Primary source MGI:MGI:107335

See related Ensembl: ENSMUSG00000002384

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 Op3

Summary This gene encodes a secreted ligand of the TGF-beta (transforming growth factor-beta) superfamily of proteins. Ligands of this family bind

various TGF-beta receptors leading to recruitment and activation of SMAD family transcription factors that regulate gene expression. The encoded preproprotein is proteolytically processed to generate each subunit of the disulfide-linked homodimer. The encoded protein may play a role in the generation of primordial germ cells, and has been shown to stimulate thermogenesis in brown adipose tissue. Male mice lacking a functional copy of this gene exhibit variable degrees of germ-cell deficiency. Homozygous knockout mice of both sexes exhibit impaired thermogenesis and reduced metabolic rate, resulting in weight gain. This gene may have arose from a gene duplication event and its gene

duplicate is also present on chromosome 4. [provided by RefSeq, Jul 2016]

Expression Biased expression in small intestine adult (RPKM 3.9), large intestine adult (RPKM 1.9) and 8 other tissuesSee more

Orthologs <u>human</u> all

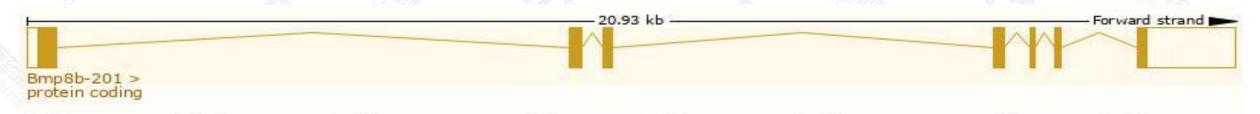
Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

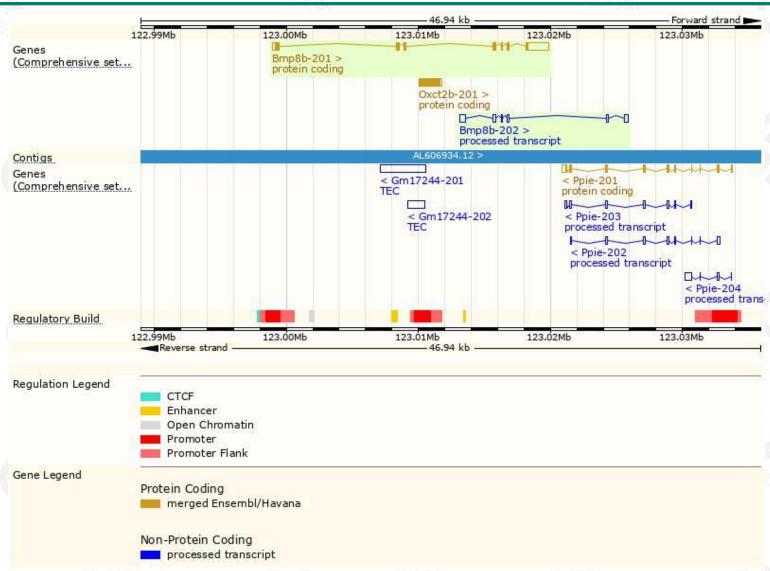
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bmp8b-201	ENSMUST00000002457.1	2940	399aa	Protein coding	CCDS18608	P55105	TSL:1 GENCODE basic APPRIS P1
Bmp8b-202	ENSMUST00000151850.1	1326	No protein	Processed transcript		(-)	TSL:1

The strategy is based on the design of *Bmp8b-201* transcript, the transcription is shown below:



Genomic location distribution





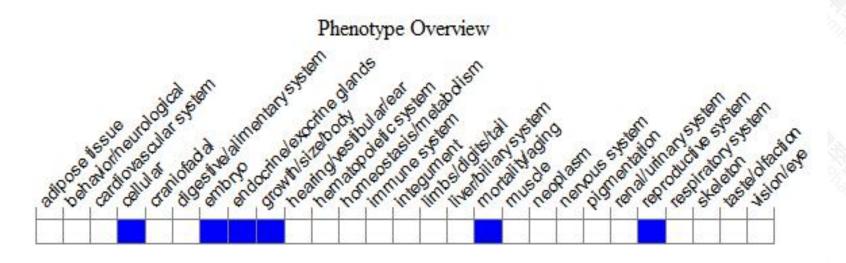
Protein domain





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,incidence of lethality among homozygous null mutants is variable depending on genetic background and due to allantoic and embryonic abnormalities. Heterozygous and surviving homozygous males exhibit varying degrees of germ cell deficiency and infertility, also background dependent.



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

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