

***Bmp2* Cas9-CKO Strategy**

Designer:

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Design Date:

2019-7-17

Project Overview

Project Name

Bmp2

Project type

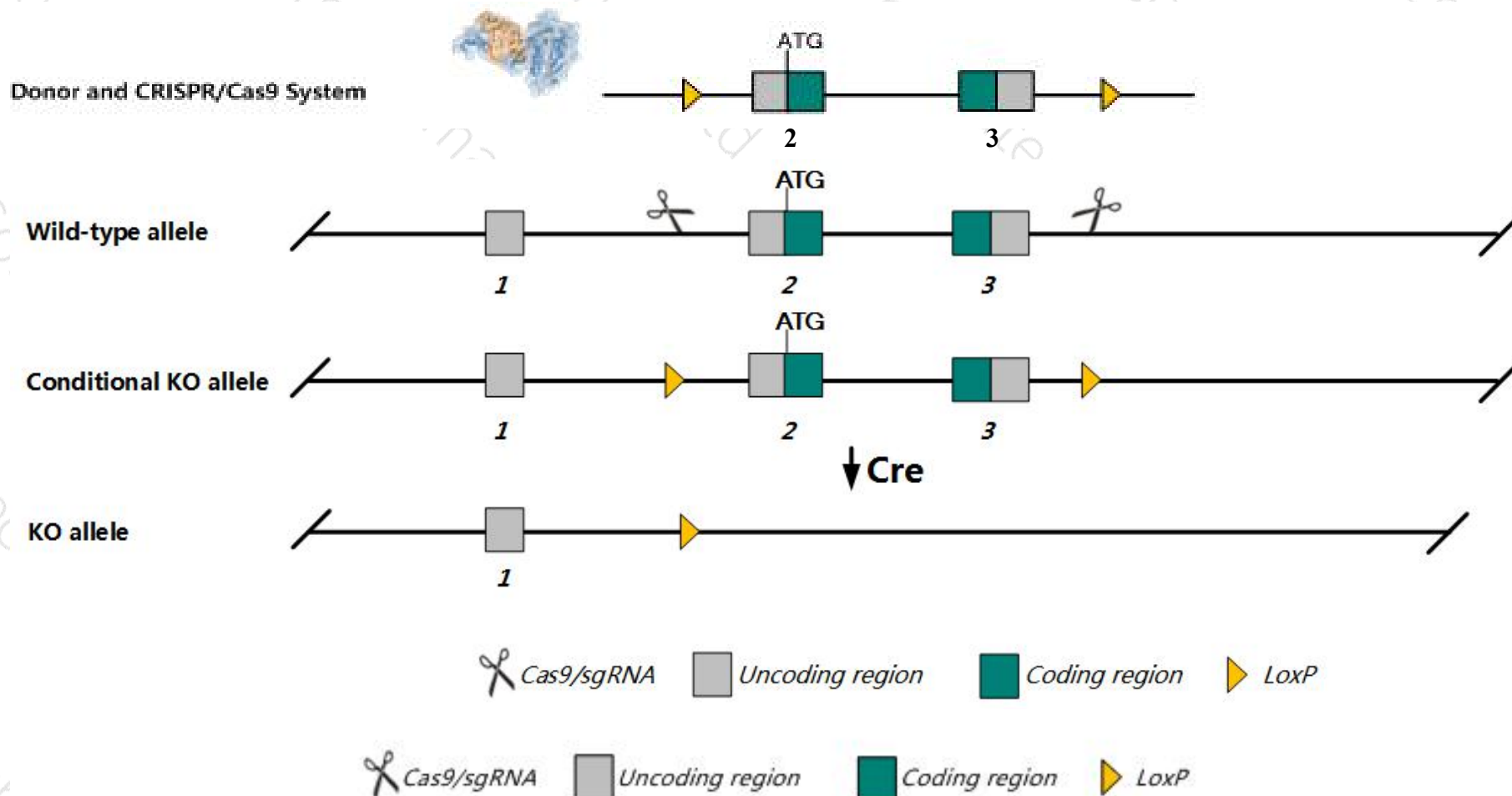
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Bmp2* gene. The schematic diagram is as follows:



- The *Bmp2* gene has 1 transcript. According to the structure of *Bmp2* gene, exon2-exon3 of *Bmp2-201* (ENSMUST00000028836.6) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Bmp2* 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, Homozygous null mutants die at embryonic day 7.5-9 with failure of the proamniotic canal to close and abnormal development of the heart in the exocoelomic cavity.
- The *Bmp2* gene is located on the Chr2. 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)

Bmp2 bone morphogenetic protein 2 [Mus musculus (house mouse)]

Gene ID: 12156, updated on 31-Jan-2019

Summary



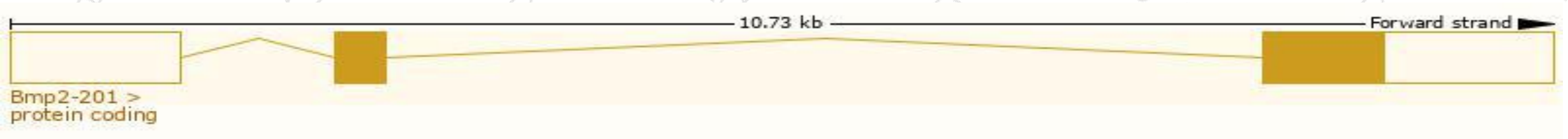
Official Symbol	Bmp2 provided by MGI
Official Full Name	bone morphogenetic protein 2 provided by MGI
Primary source	MGI:MGI:88177
See related	Ensembl:ENSMUSG000000027358
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI467020, Bmp2a
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, which plays a role in bone and cartilage development. While a homozygous knockout mouse for this gene exhibits embryonic lethality, conditional knockout mice have defects in bone, cartilage and heart development. [provided by RefSeq, Jul 2016]
Expression	Biased expression in colon adult (RPKM 19.7), stomach adult (RPKM 14.3) and 13 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

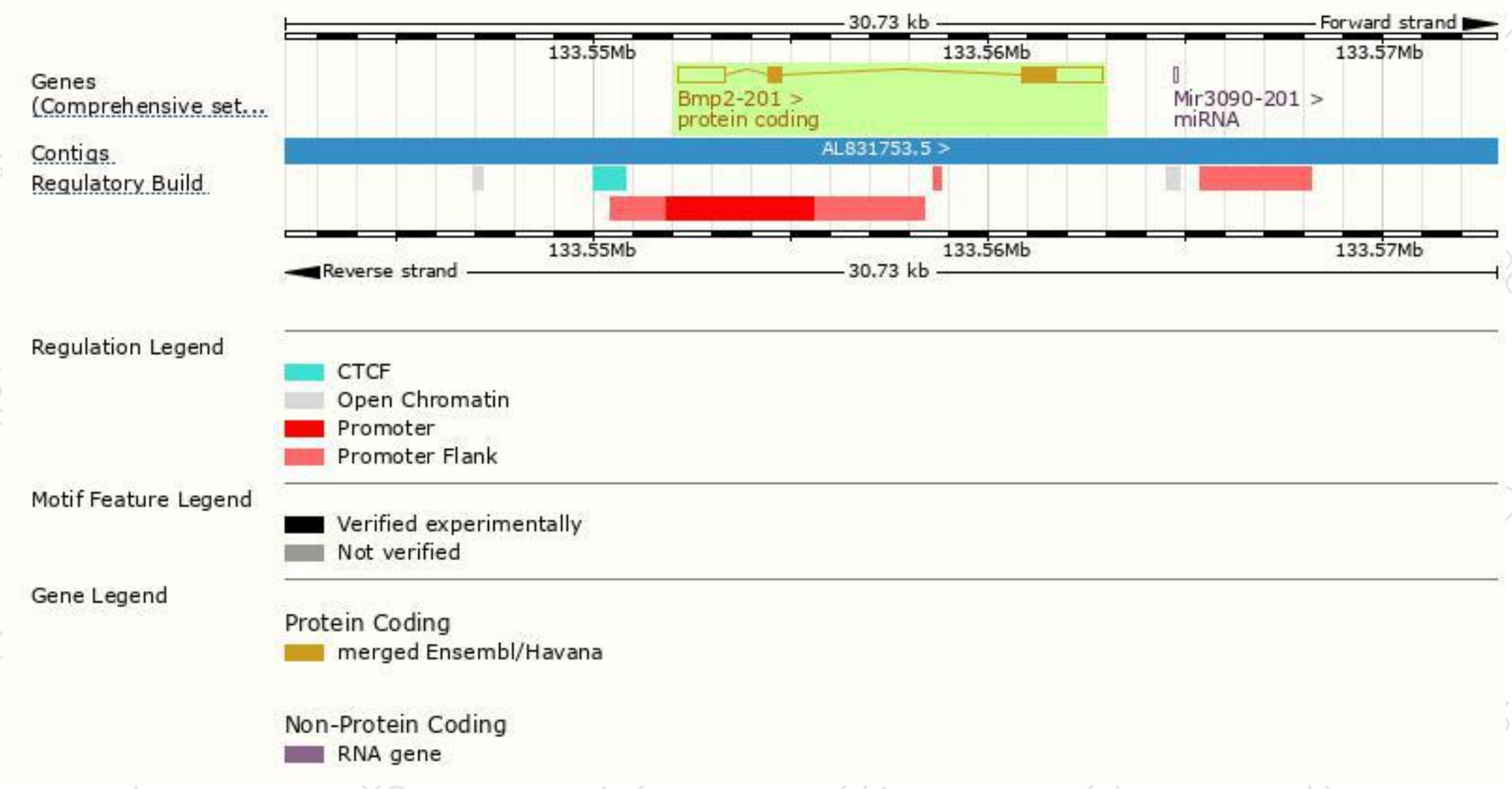
The gene has 1 transcript, and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bmp2-201	ENSMUST00000028836.6	3555	394aa	Protein coding	CCDS16782	P21274	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of *Bmp2-201* transcript, The transcription is shown below



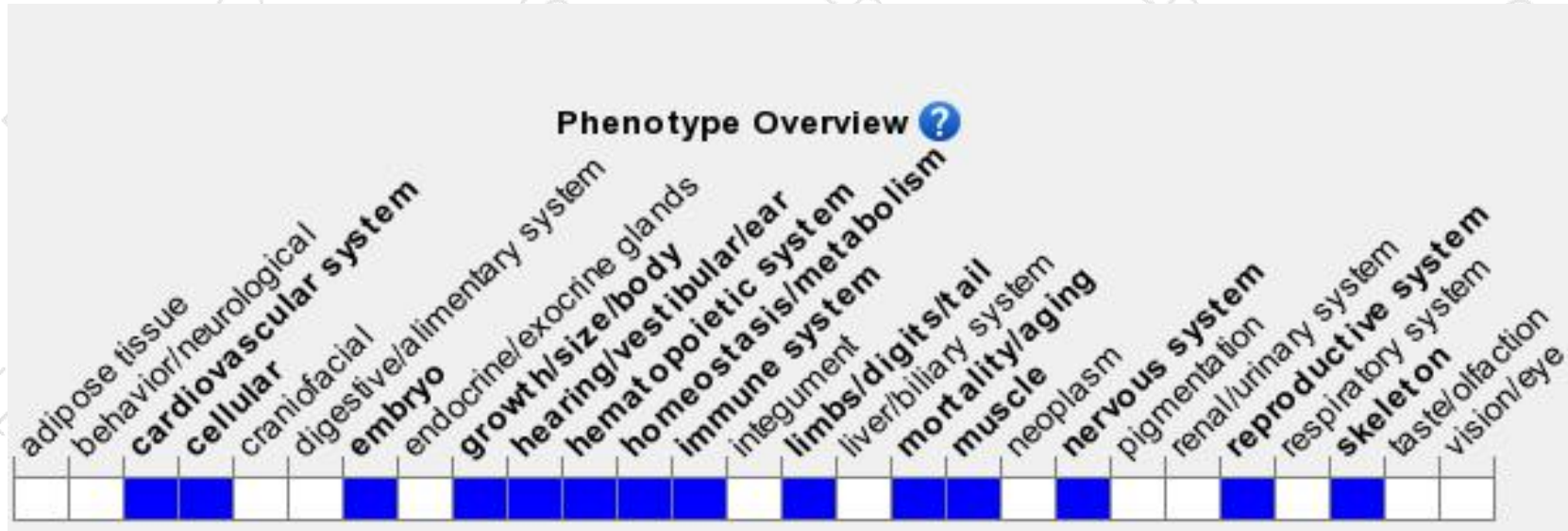
Genomic location distribution



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 die at embryonic day 7.5-9 with failure of the proamniotic canal to close and abnormal development of the heart in the exocoelomic cavity.

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

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