# *Ibsp* Cas9-CKO *Strategy*

**Designer:** **Ruirui Zhang**
**Reviewer:** **Huimin Su**

**Design Date:** **2020-3-24**

# Project Overview

| | |
|---|---|
| **Project Name** | ***Ibsp*** |
| **Project type** | **Cas9-CKO** |
| **Strain background** | **C57BL/6J** |

# Conditional Knockout strategy

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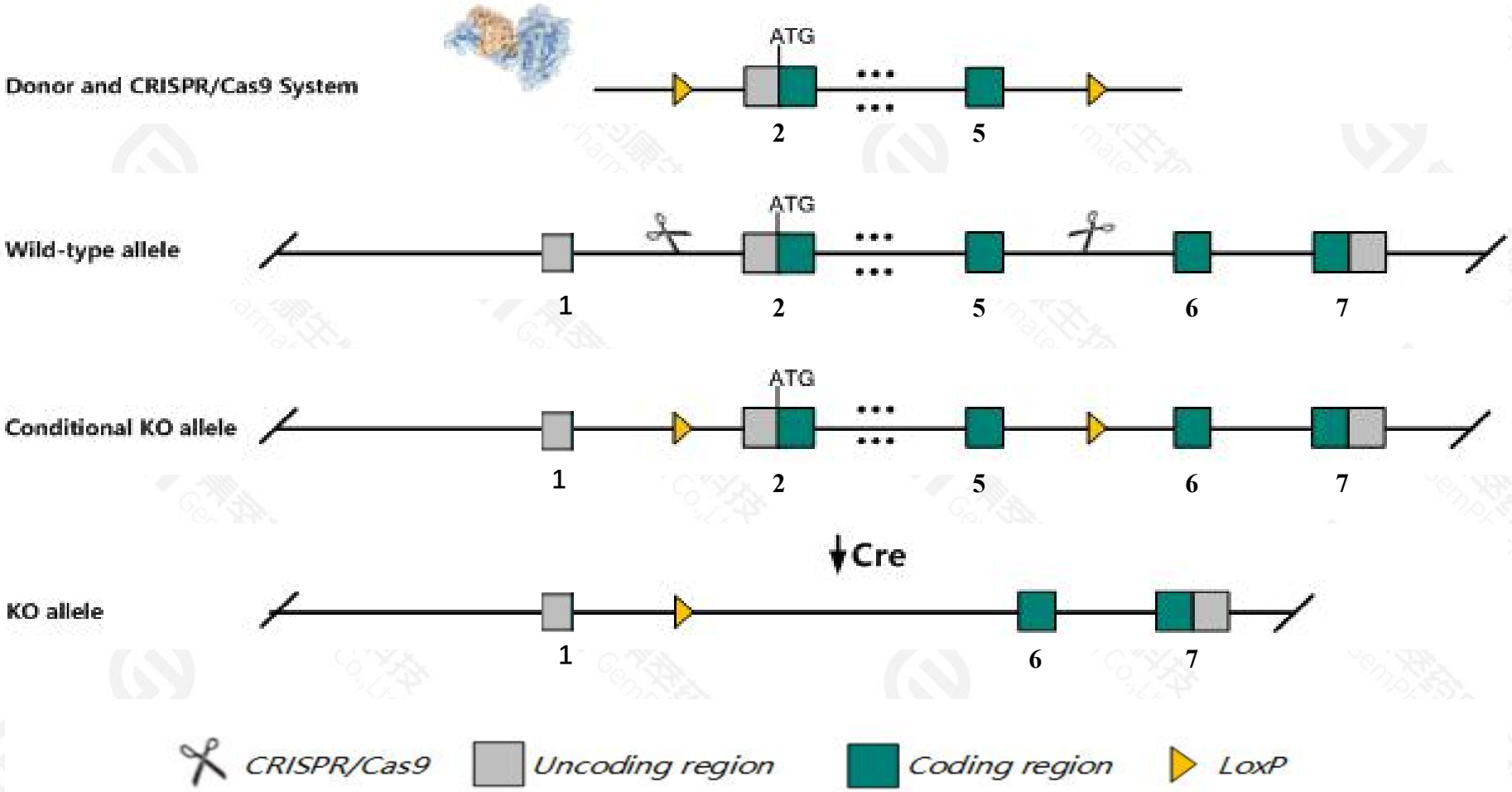
This model will use CRISPR/Cas9 technology to edit the *Ibsp* gene. The schematic diagram is as follows:



# Technical routes

- The *Ibsp* gene has 1 transcript. According to the structure of *Ibsp* gene, exon2~exon5 of *Ibsp*-201 (ENSMUST00000031246.8)transcript is recommended as the knockout region.The region contains the start codon ATG.Knock out the region will result in disruption of protein function.
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- In this project we use CRISPR/Cas9 technology to modify *Ibsp* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA and Donor were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

- The flox mice was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.

# Notice

- According to the existing MGI data，mice homozygous for a knock-out allele show reduced body weight/size, delayed long bone growth and mineralization with low bone turn over due to reduced osteoclast formation, delayed intramembranous ossification, progressive periodontal breakdown, and severe alveolar and mandibular bone loss.
- The *Ibsp* gene is located on the Chr5. 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

# Gene information (NCBI)

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## Ibsp integrin binding sialoprotein [ *Mus musculus* (house mouse) ]

Gene ID: 15891, updated on 13-Mar-2020

### Summary

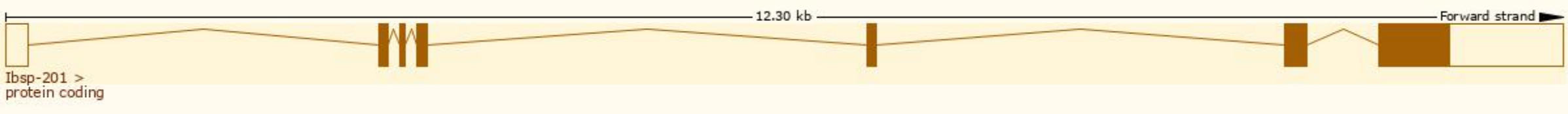
| | |
|---|---|
| **Official Symbol** | Ibsp provided by MGI |
| **Official Full Name** | integrin binding sialoprotein provided by MGI |
| **Primary source** | MGI:MGI:96389 |
| **See related** | Ensembl:ENSMUSG00000029306 |
| **Gene type** | protein coding |
| **RefSeq status** | VALIDATED |
| **Organism** | Mus musculus |
| **Lineage** | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus |
| **Also known as** | BSP; Bsp2; BSPII; BSP II |
| **Expression** | Biased expression in limb E14.5 (RPKM 37.3), CNS E14 (RPKM 11.6) and 2 other tissues See more |
| **Orthologs** | human all |

# Transcript information （Ensembl）

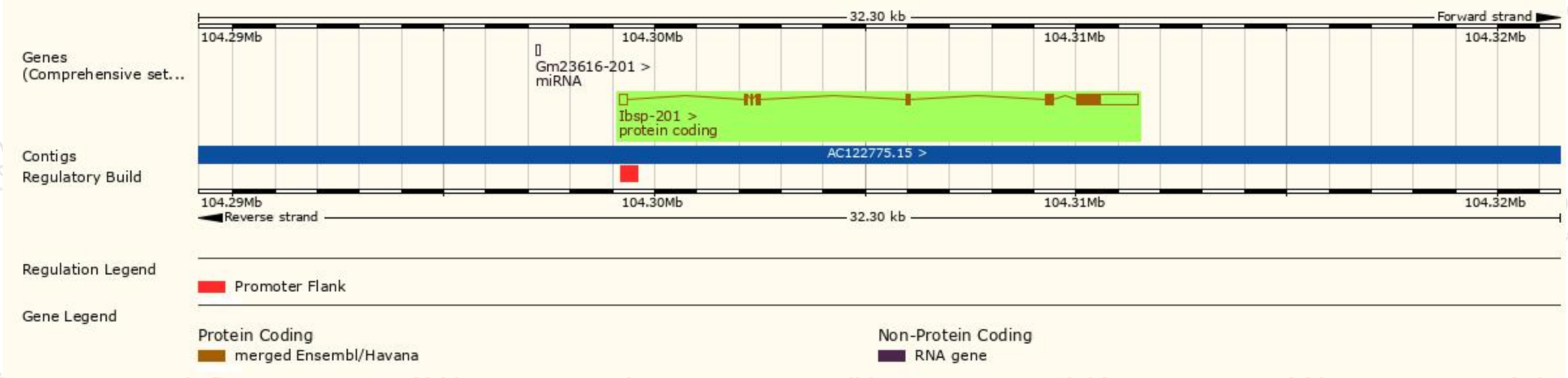
The gene has 1 transcript, and all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| Ibsp-201 | ENSMUST00000031246.8 | 2068 | 324aa | Protein coding | CCDS19485 | Q61711 | TSL:1 GENCODE basic APPRIS P1 |

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

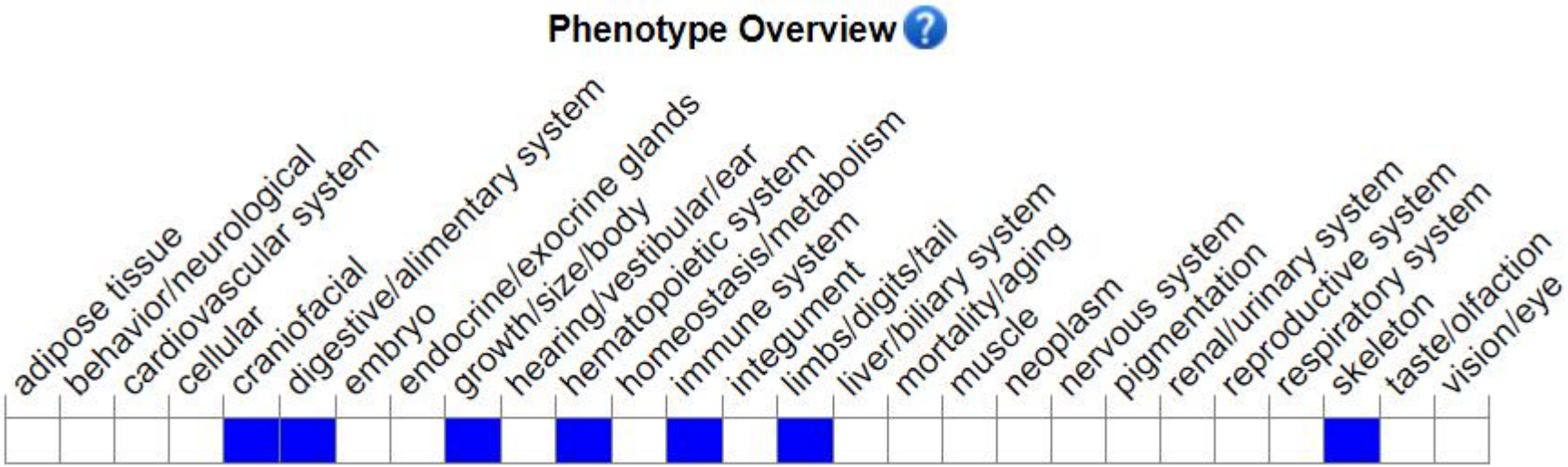


# Genomic location distribution



# 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, mice homozygous for a knock-out allele show reduced body weight/size, delayed long bone growth and mineralization with low bone turn over due to reduced osteoclast formation, delayed intramembranous ossification, progressive periodontal breakdown, and severe alveolar and mandibular bone loss.

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

Tel: 025-5864 1534

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GemPharmatech Co.,Ltd