

***Bend3* Cas9-CKO Strategy**

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Project Overview

Project Name

Bend3

Project type

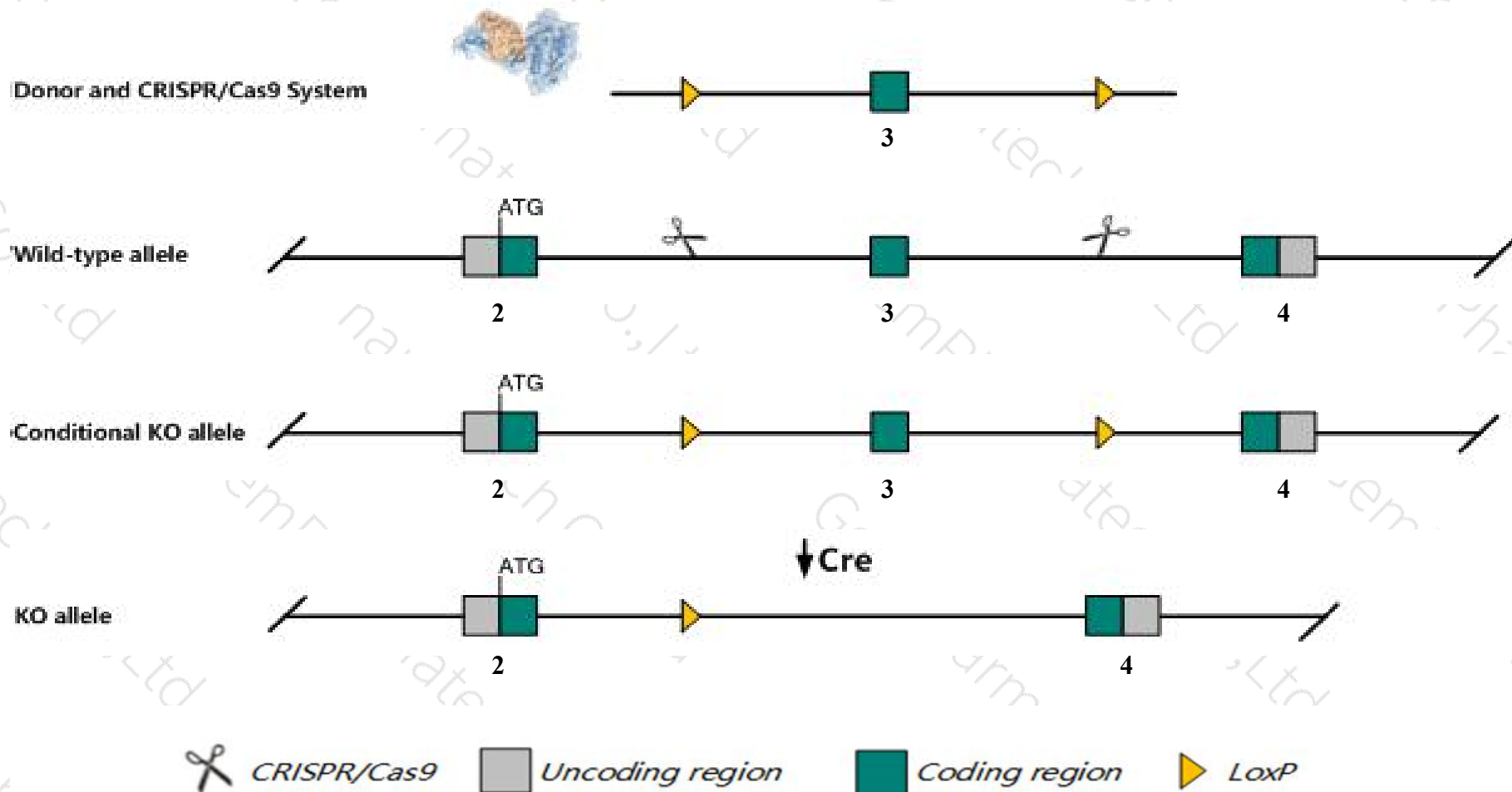
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Bend3* gene. The schematic diagram is as follows:



Technical routes

- The *Bend3* gene has 4 transcripts. According to the structure of *Bend3* gene, exon3 of *Bend3*-202(ENSMUST00000167488.8) transcript is recommended as the knockout region. The region contains 197bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Bend3* 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.

- The Intron2 and Intron3 are only 456bp and 15504bp, loxp insertion may affect mRNA splicing.
- The *Bend3* gene is located on the Chr10. 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)

Bend3 BEN domain containing 3 [Mus musculus (house mouse)]

Gene ID: 331623, updated on 13-Mar-2020

Summary



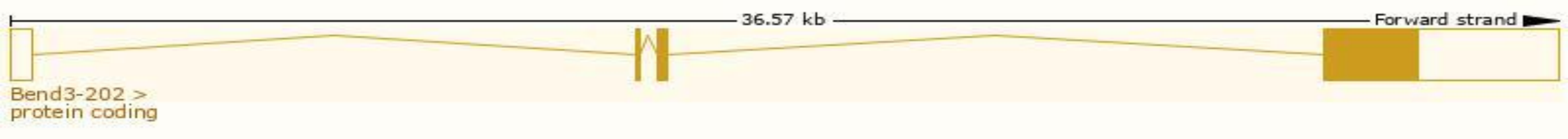
Official Symbol	Bend3 provided by MGI
Official Full Name	BEN domain containing 3 provided by MGI
Primary source	MGI:MGI:2677212
See related	Ensembl:ENSMUSG00000038214
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	mKIAA1553
Expression	Ubiquitous expression in CNS E11.5 (RPKM 3.0), limb E14.5 (RPKM 2.1) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

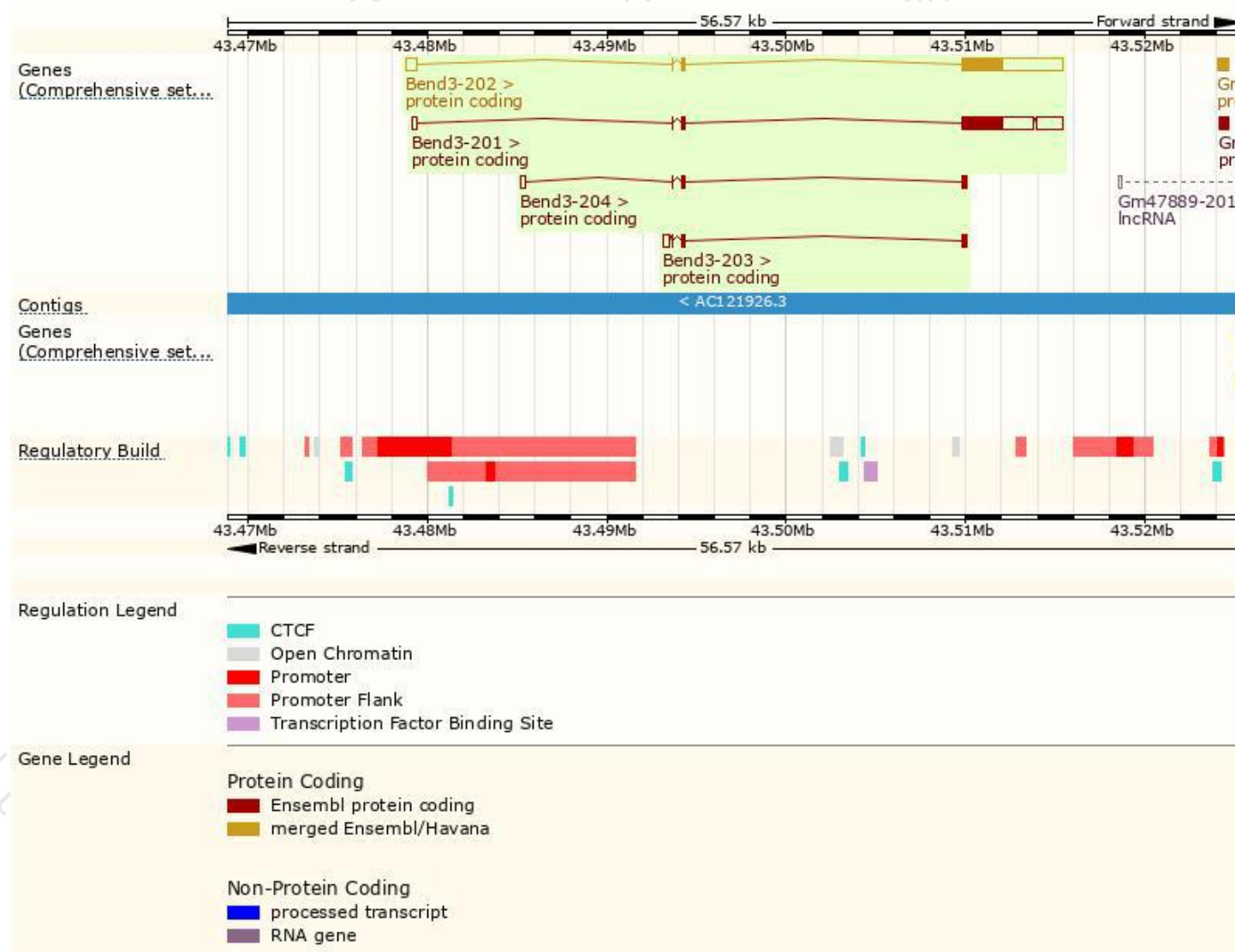
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bend3-202	ENSMUST00000167488.8	6337	825aa	Protein coding	CCDS35892	Q6PAL0	TSL:2 GENCODE basic APPRIS P1
Bend3-201	ENSMUST00000040147.7	5882	825aa	Protein coding	CCDS35892	Q6PAL0	TSL:1 GENCODE basic APPRIS P1
Bend3-203	ENSMUST00000214116.1	746	144aa	Protein coding	-	A0A1L1SSB5	CDS 3' incomplete TSL:2
Bend3-204	ENSMUST00000216679.1	670	159aa	Protein coding	-	A0A1L1SU50	CDS 3' incomplete TSL:5

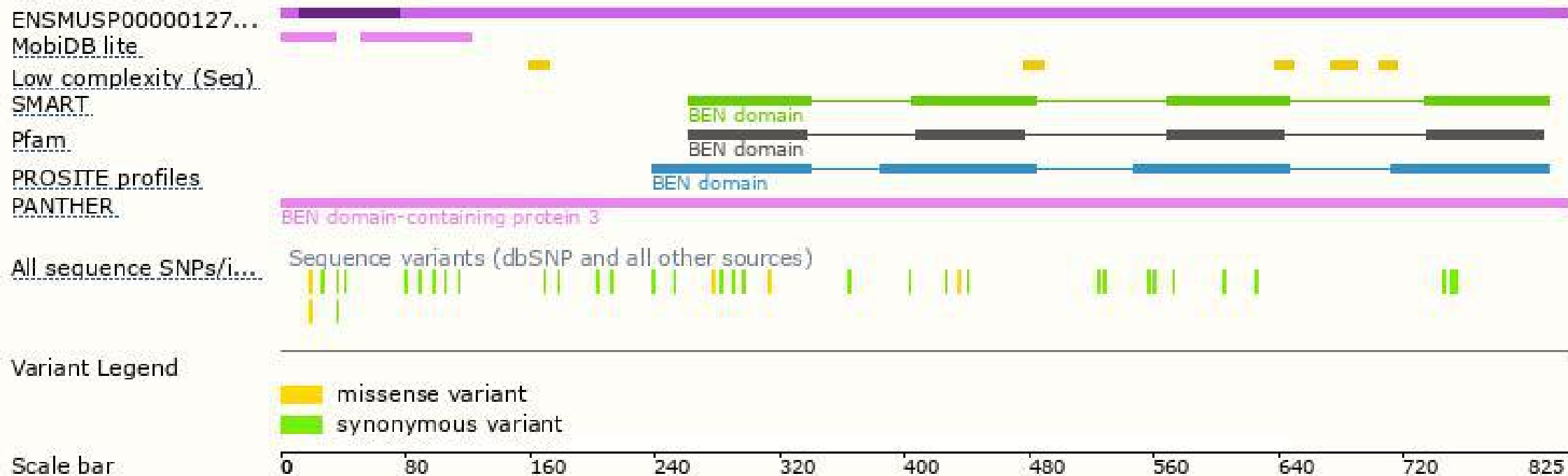
The strategy is based on the design of *Bend3-202* transcript,the transcription is shown below:



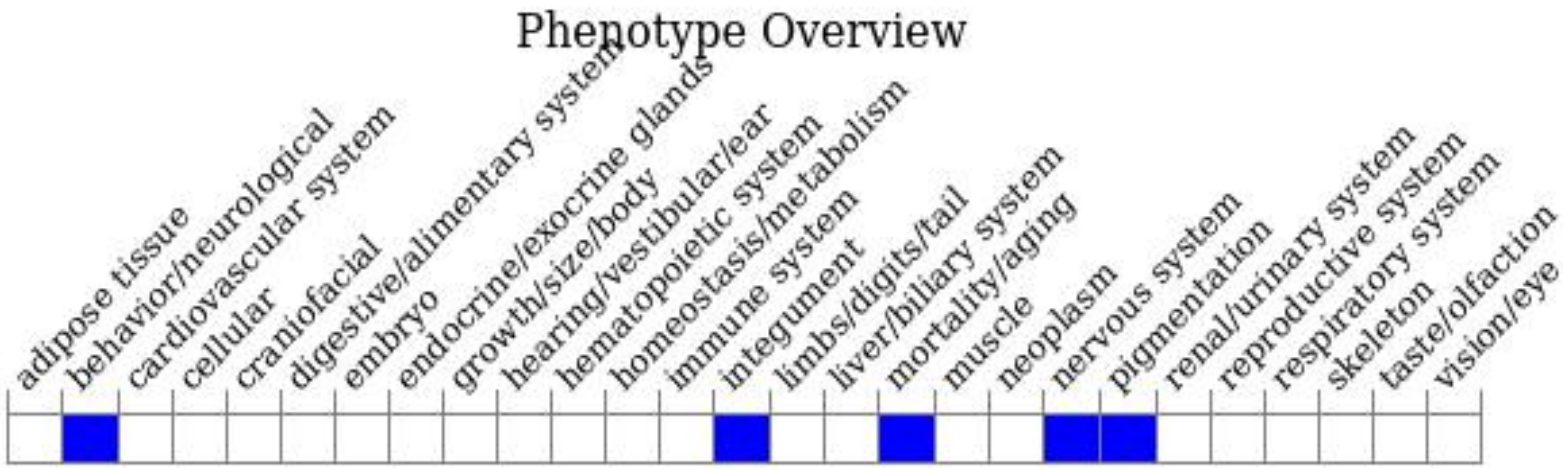
Genomic location distribution



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/>).

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

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