Bend5 Cas9-KO Strategy

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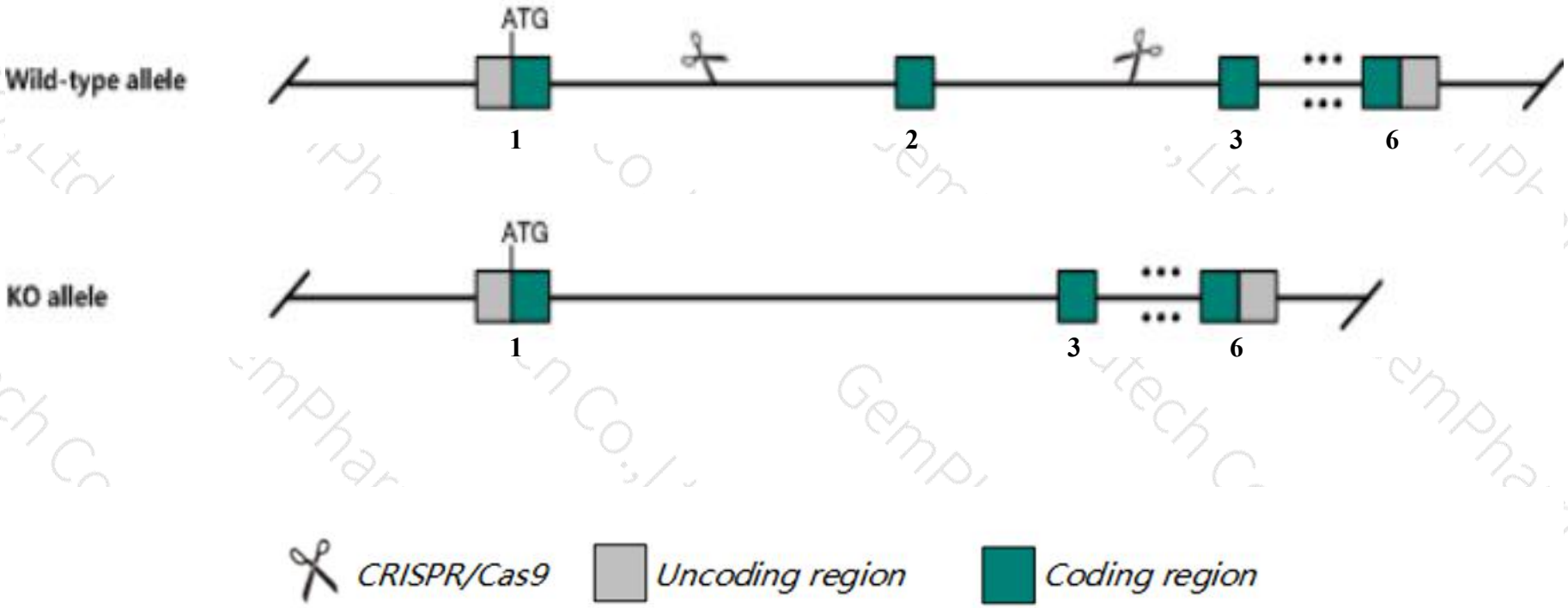
Design Date: 2020-11-19

Project Overview

Project Name	***Bend5***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Bend5* gene. The schematic diagram is as follows:



Technical routes

➢ The *Bend5* gene has 6 transcripts. According to the structure of *Bend5* gene, exon2 of *Bend5-201*(ENSMUST00000030274.6) transcript is recommended as the knockout region.The region contains 134bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Bend5* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ Transcript *Bend5-206* may not be affected.

➢ The KO region contains functional region of the *Agbl4* gene.Knockout the region may affect the function of *Agbl4* gene.

➢ The *Bend5* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

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Bend5 BEN domain containing 5 [Mus musculus (house mouse)]

Gene ID: 67621, updated on 13-Mar-2020

Summary

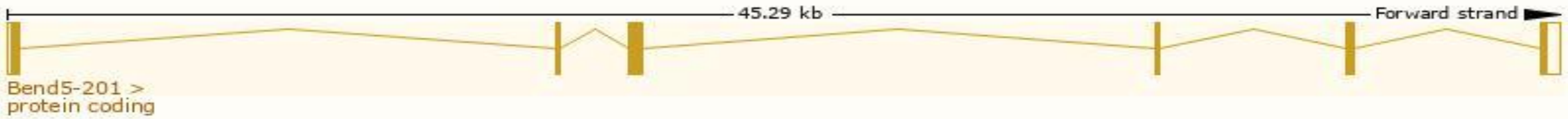
Official Symbol	Bend5 provided by MGI
Official Full Name	BEN domain containing 5 provided byMGI
Primary source	MGI:MGI:1914871
See related	Ensembl:ENSMUSG00000028545
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2310026E23Rik
Expression	Broad expression in CNS E14 (RPKM 7.7), whole brain E14.5 (RPKM 7.2) and 24 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

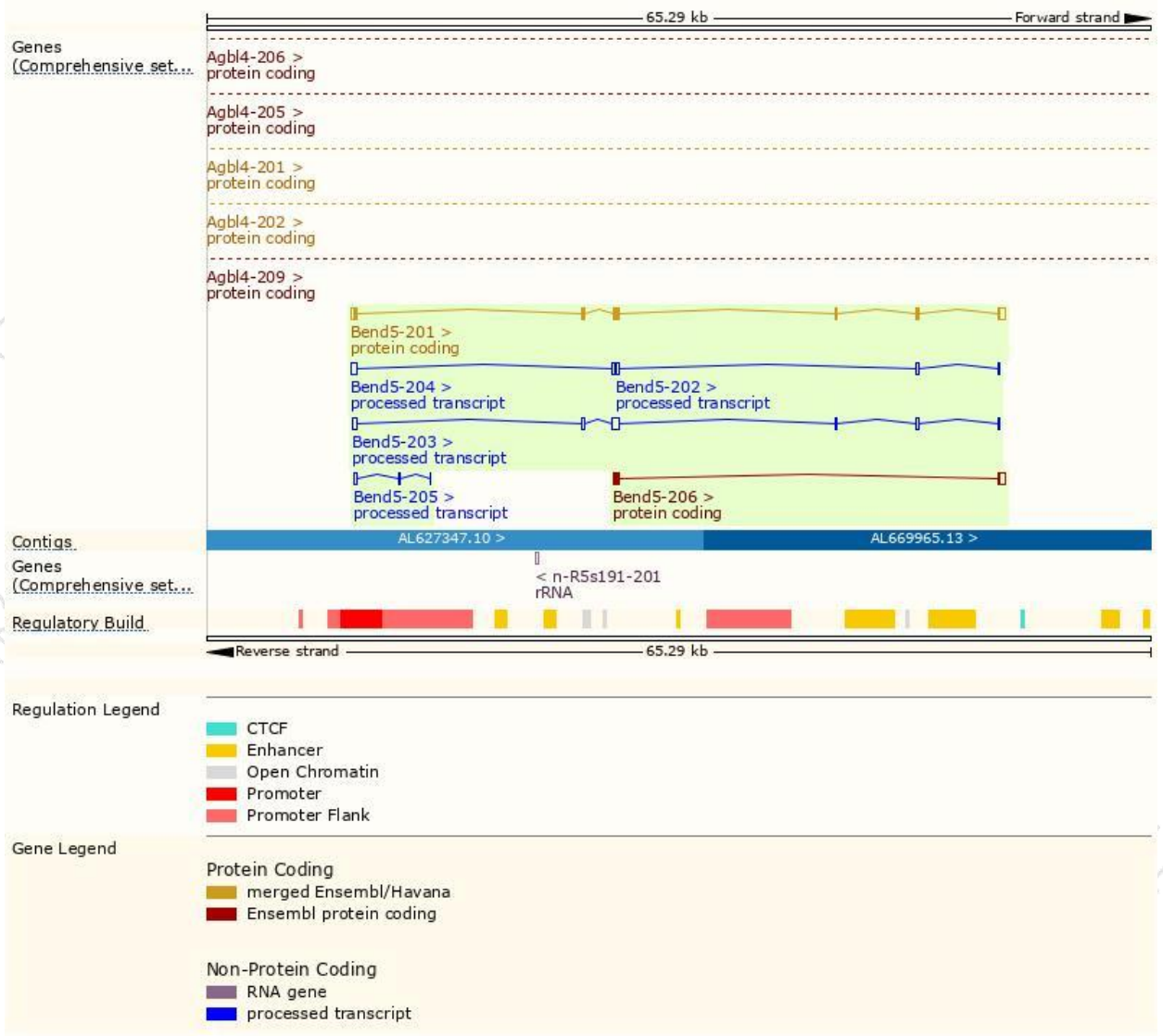
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bend5-201	ENSMUST00000030274.6	1826	421aa	Protein coding	CCDS18472	Q8C6D4	TSL:1 GENCODE basic APPRIS P1
Bend5-206	ENSMUST00000139876.1	904	171aa	Protein coding	-	F6Q2G7	CDS 5' incomplete TSL:2
Bend5-203	ENSMUST00000129760.7	1448	No protein	Processed transcript	-	-	TSL:2
Bend5-202	ENSMUST00000123667.1	627	No protein	Processed transcript	-	-	TSL:3
Bend5-204	ENSMUST00000130523.1	514	No protein	Processed transcript	-	-	TSL:5
Bend5-205	ENSMUST00000138394.1	391	No protein	Processed transcript	-	-	TSL:5

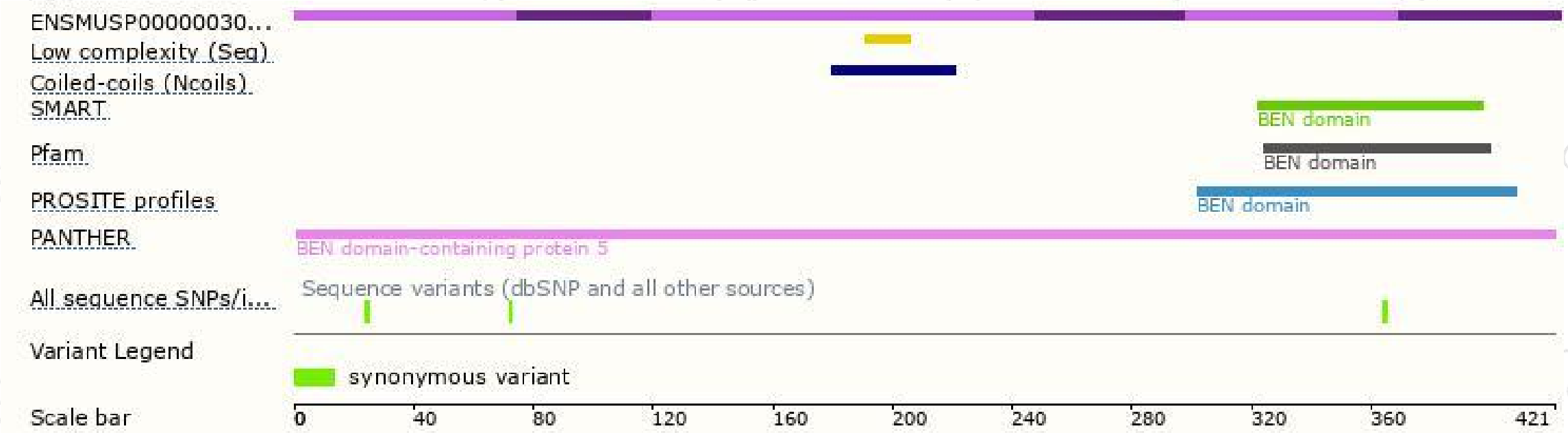
The strategy is based on the design of *Bend5-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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