Bbs2 Cas9-KO Strategy

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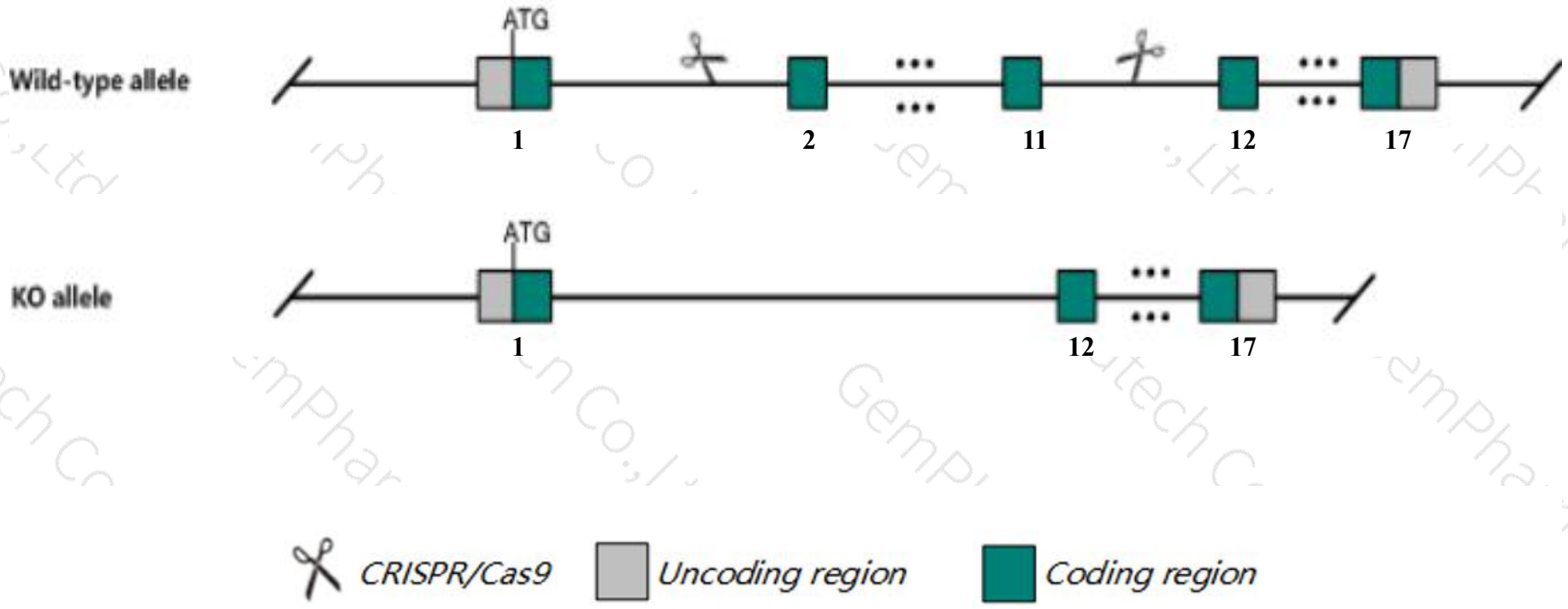
Project Overview

Project Name	***Bbs2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Bbs2* gene. The schematic diagram is as follows:



Technical routes

➢ The *Bbs2* gene has 3 transcripts. According to the structure of *Bbs2* gene, exon2-exon11 of *Bbs2-201*(ENSMUST00000034206.5) transcript is recommended as the knockout region.The region contains 1280bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Bbs2* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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

➢ According to the existing MGI data,homozygous null mice display obesity associated with polyphagia, retinopathy associated with mislocalization of rhodopsin, cilia defects, renal cysts, male sterility, abnormal brain neuroanatomy, reduced salivation and acoustic startle response, an olfactory deficit and abnormal social interaction.

➢ The *Bbs2* gene is located on the Chr8. 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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Bbs2 Bardet-Biedl syndrome 2 (human) [Mus musculus (house mouse)]

Gene ID: 67378, updated on 13-Mar-2020

Summary

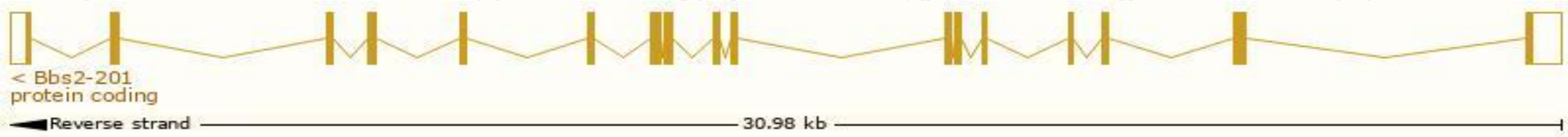
Official Symbol	Bbs2 provided by MGI
Official Full Name	Bardet-Biedl syndrome 2 (human) provided byMGI
Primary source	MGI:MGI:2135267
See related	Ensembl:ENSMUSG00000031755
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2410125H22Rik, AI447581
Expression	Ubiquitous expression in frontal lobe adult (RPKM 8.6), testis adult (RPKM 8.2) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

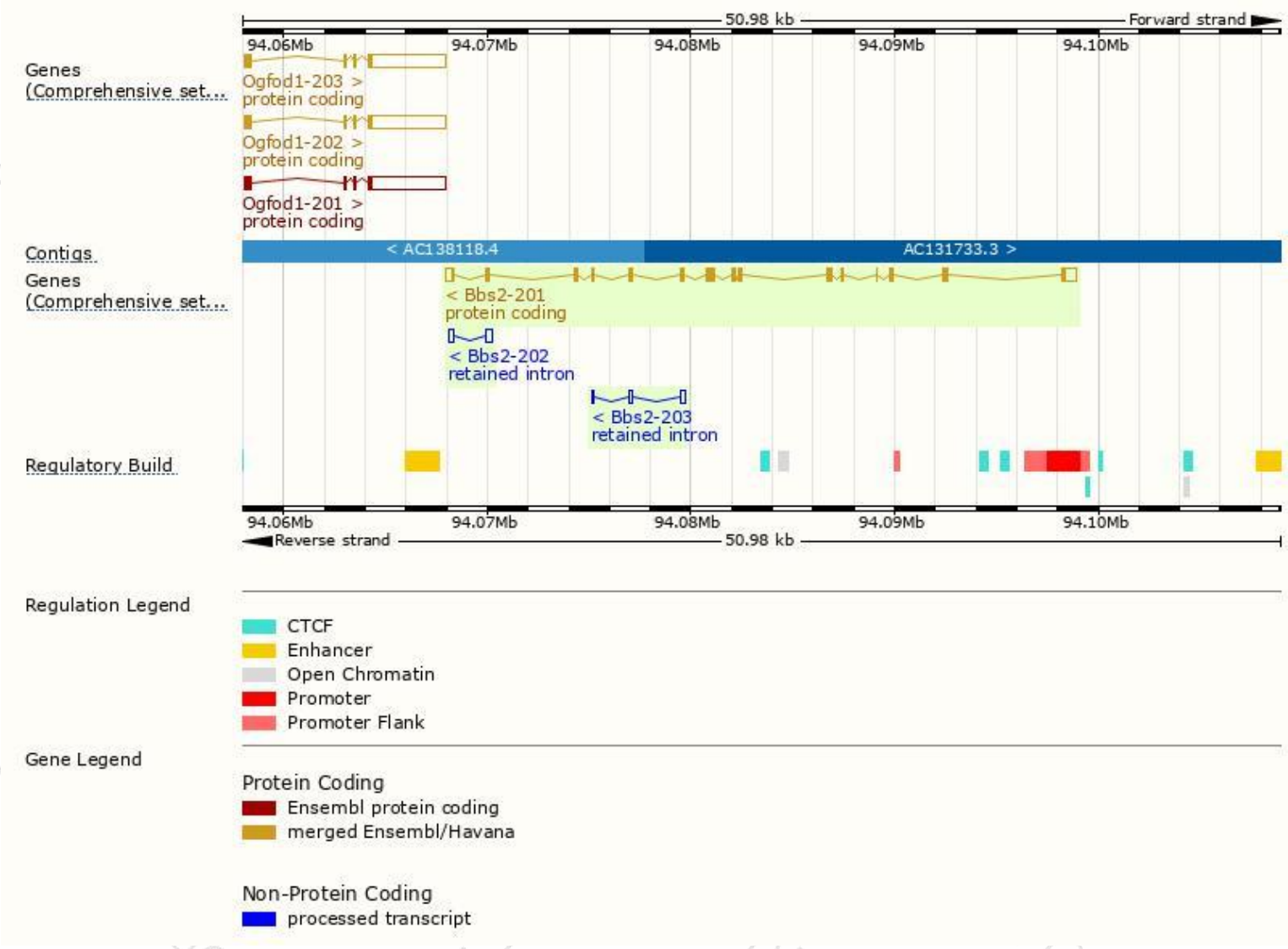
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bbs2-201	ENSMUST00000034206.5	3064	721aa	Protein coding	CCDS40434	Q9CWF6	TSL:1 GENCODE basic APPRIS P1
Bbs2-203	ENSMUST00000172347.1	522	No protein	Retained intron	-	-	TSL:2
Bbs2-202	ENSMUST00000170208.1	491	No protein	Retained intron	-	-	TSL:1

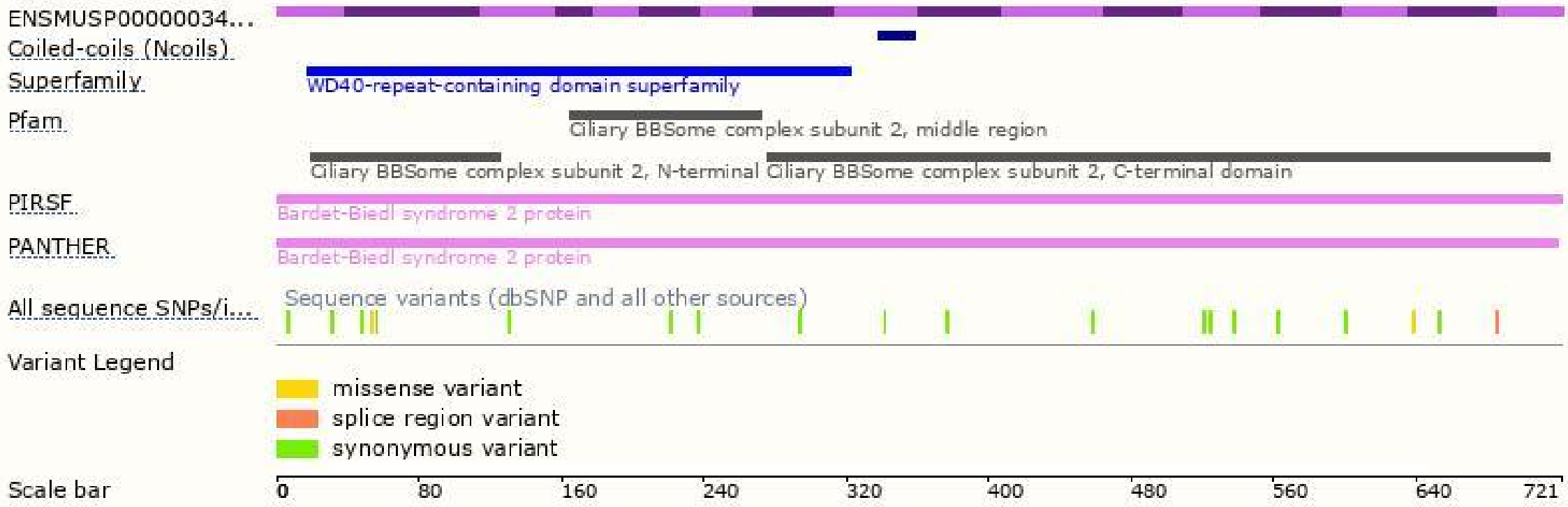
The strategy is based on the design of *Bbs2-201* transcript,the transcription is shown below:



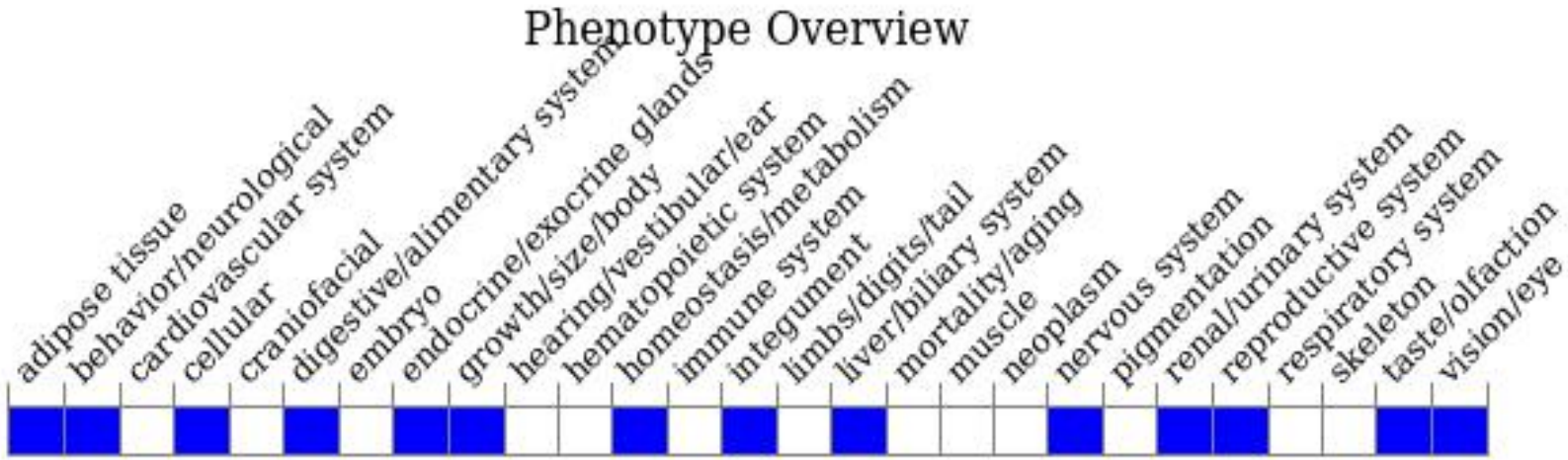
Genomic location distribution



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/).

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