Otud6b Cas9-KO *Strategy*

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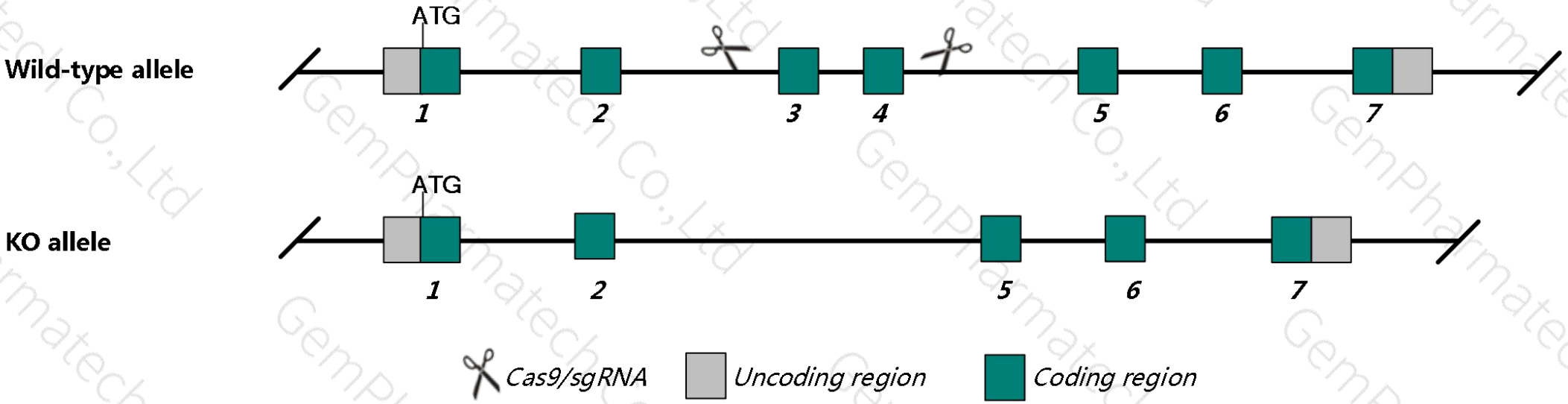
Project Overview

Project Name	***Otud6b***
Project type	**Cas9-KO**
Strain background	**C57BL/6J**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Otud6b* gene. The schematic diagram is as follows:



Technical routes

- The *Otud6b* gene has 3 transcripts. According to the structure of *Otud6b* gene, exon3-4 of *Otud6b*-203 (ENSMUST00000236953.1)transcript is recommended as the knockout region.The region contains 394bp coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Otud6b* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice

- According to the existing MGI data，mice homozygous for a knock-out allele exhibit complete perinatal lethality, decreased fetal size, and ventricular septal defects.
- The *Otud6b* 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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Otud6b OTU domain containing 6B [*Mus musculus* (house mouse)]

Gene ID: 72201, updated on 24-Oct-2019

Summary

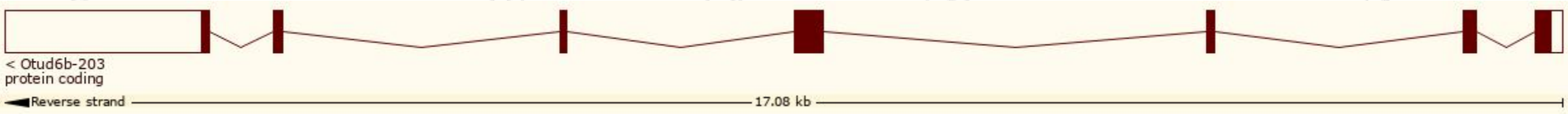
Official Symbol	Otud6b provided by MGI
Official Full Name	OTU domain containing 6B provided by MGI
Primary source	MGI:MGI:1919451
See related	Ensembl:ENSMUSG00000040550
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AU015433; 2600013N14Rik
Expression	Ubiquitous expression in bladder adult (RPKM 13.1), CNS E11.5 (RPKM 11.8) and 27 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

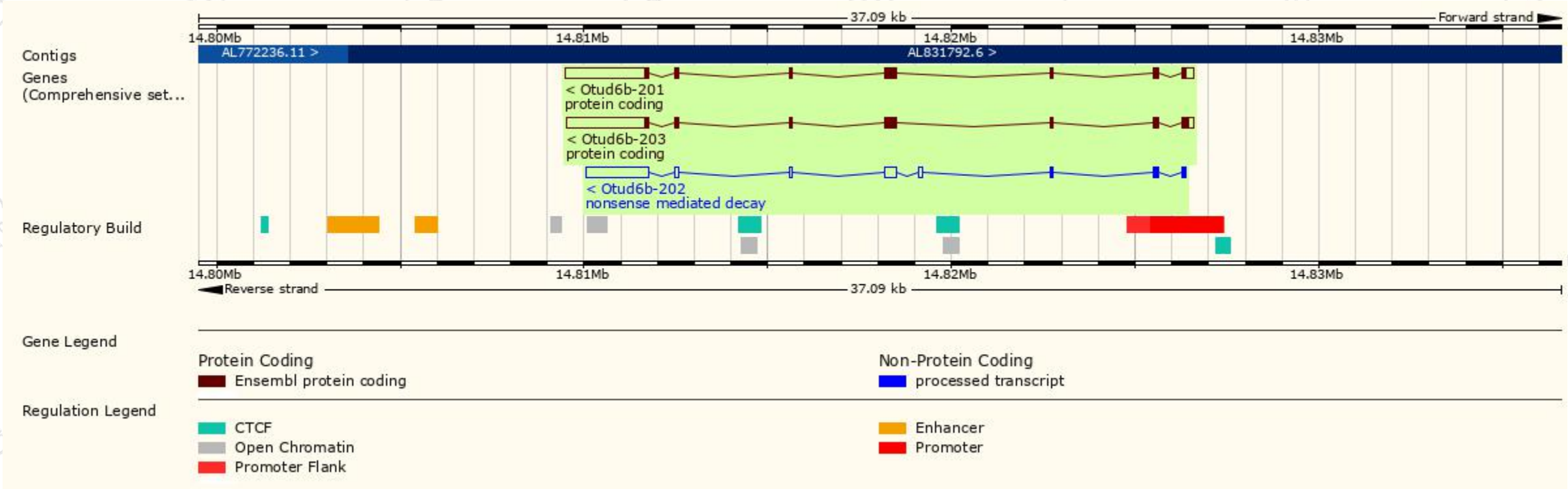
The gene has 3 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Otud6b-201	ENSMUST00000117268.8	3266	294aa	Protein coding	-	A0A0A0MQF5 Q8K2H2	TSL:1 GENCODE basic
Otud6b-202	ENSMUST00000151012.2	2647	110aa	Nonsense mediated decay	-	F6VDQ9	TSL:1
Otud6b-203	ENSMUST00000236953.1	3257	325aa	Protein coding	CCDS17980	A0A0A0MQF5	GENCODE basic APPRIS P1

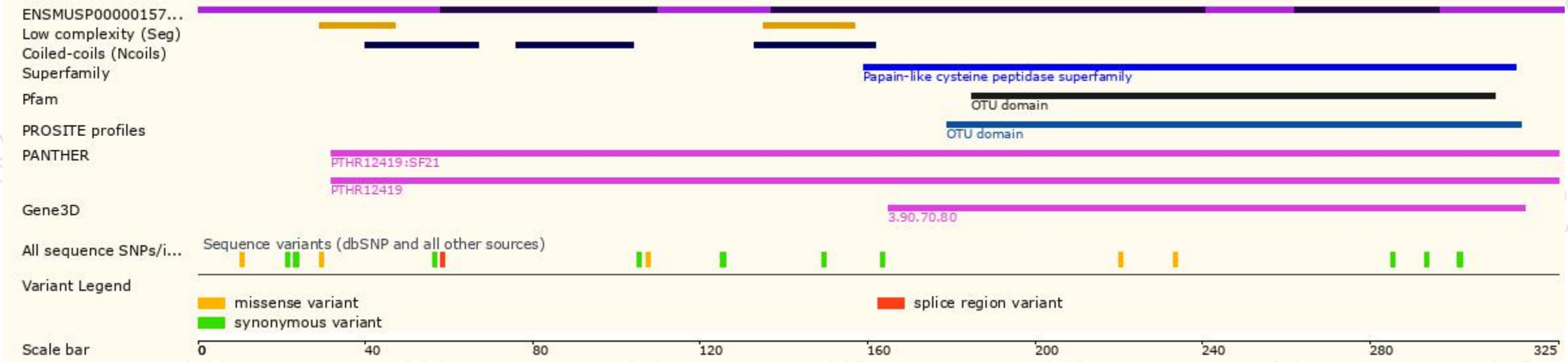
The strategy is based on the design of *Otud6b-203* transcript, the transcription is shown below：



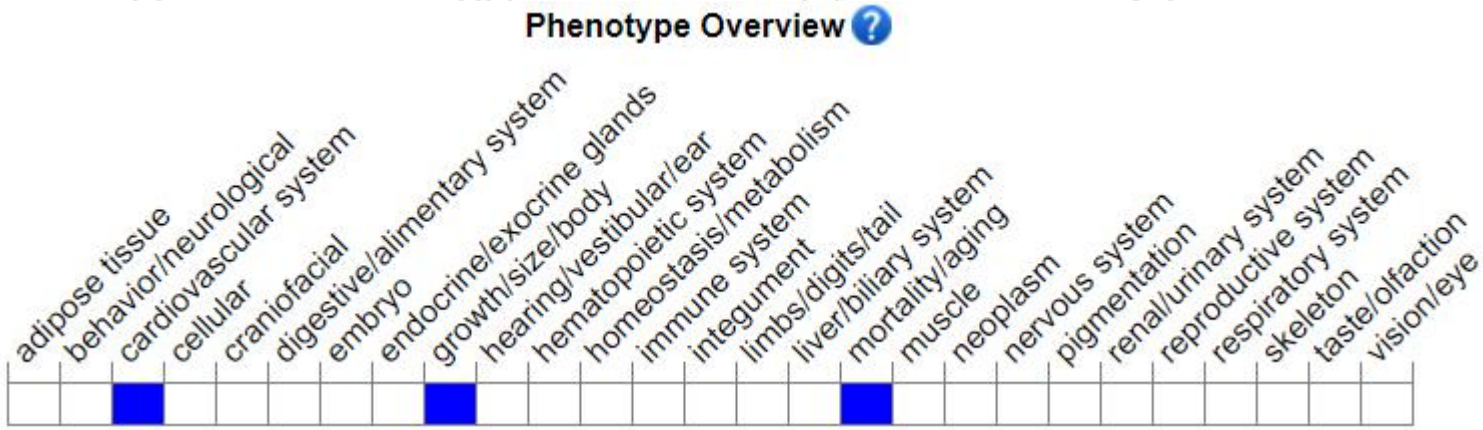
Genomic location (Ensembl)



Protein domain (Ensembl)



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 exhibit complete perinatal lethality, decreased fetal size, and ventricular septal defects.

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

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