Crybb1 Cas9-KO *Strategy*

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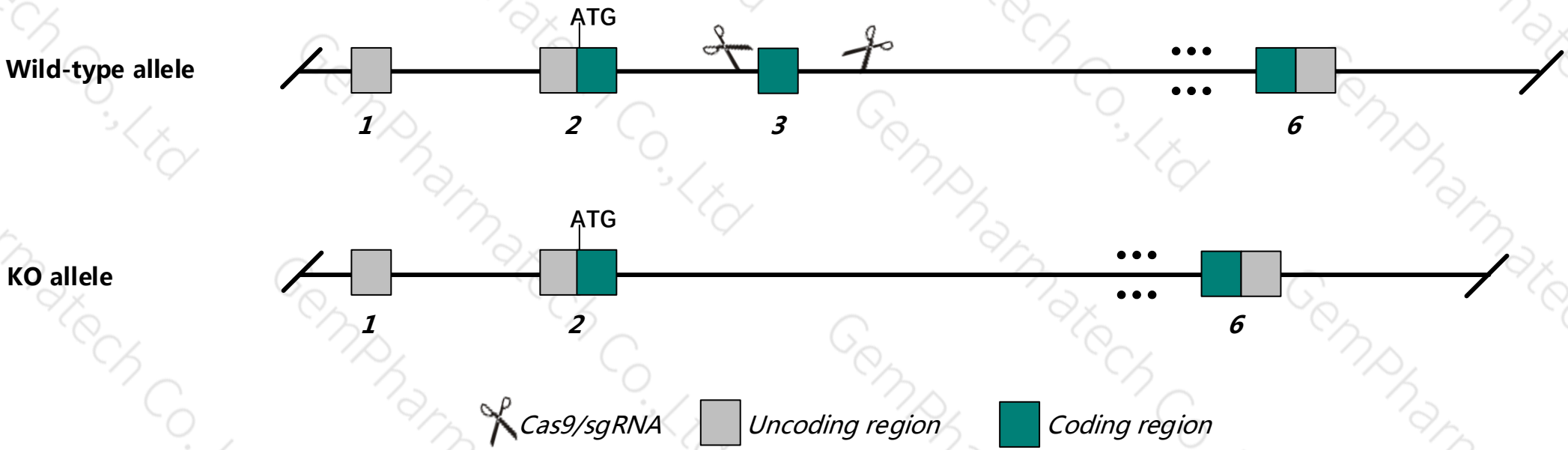
Project Overview

Project Name	***Crybb1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Crybb1* gene. The schematic diagram is as follows:



Technical routes

- The *Crybb1* gene has 3 transcripts.According to the structure of *Crybb1* gene, exon3 of *Crybb1*-201 (
- ENSMUST00000031286.12) transcript is recommended as the knockout region.The region contains 119bp coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Crybb1* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9, sgRNA 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.

Notice

- The *Crybb1* 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 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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Crybb1 crystallin, beta B1 [*Mus musculus* (house mouse)]

Gene ID: 12960, updated on 12-Aug-2019

Summary

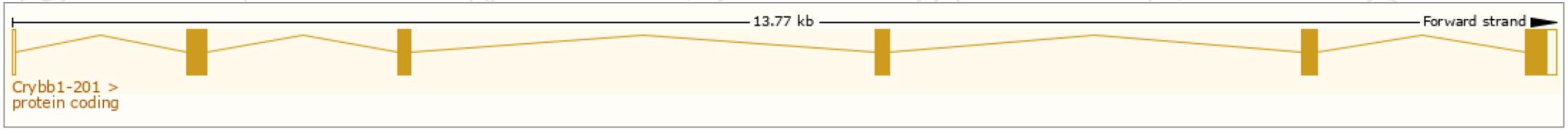
Official Symbol	Crybb1 provided by MGI
Official Full Name	crystallin, beta B1 provided by MGI
Primary source	MGI:MGI:104992
See related	Ensembl:ENSMUSG00000029343
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BB1CRY; 3110006K12Rik
Summary	This gene encodes a member of the crystallin family of proteins that contribute to the transparency and refractive properties of the ocular lens. The encoded protein associates with other beta crystallin proteins to form dimers, tetramers and other higher-order complexes. This gene is located adjacent to a related crystallin gene on chromosome 5. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Aug 2015]
Expression	Biased expression in genital fat pad adult (RPKM 1.8), ovary adult (RPKM 1.2) and 10 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

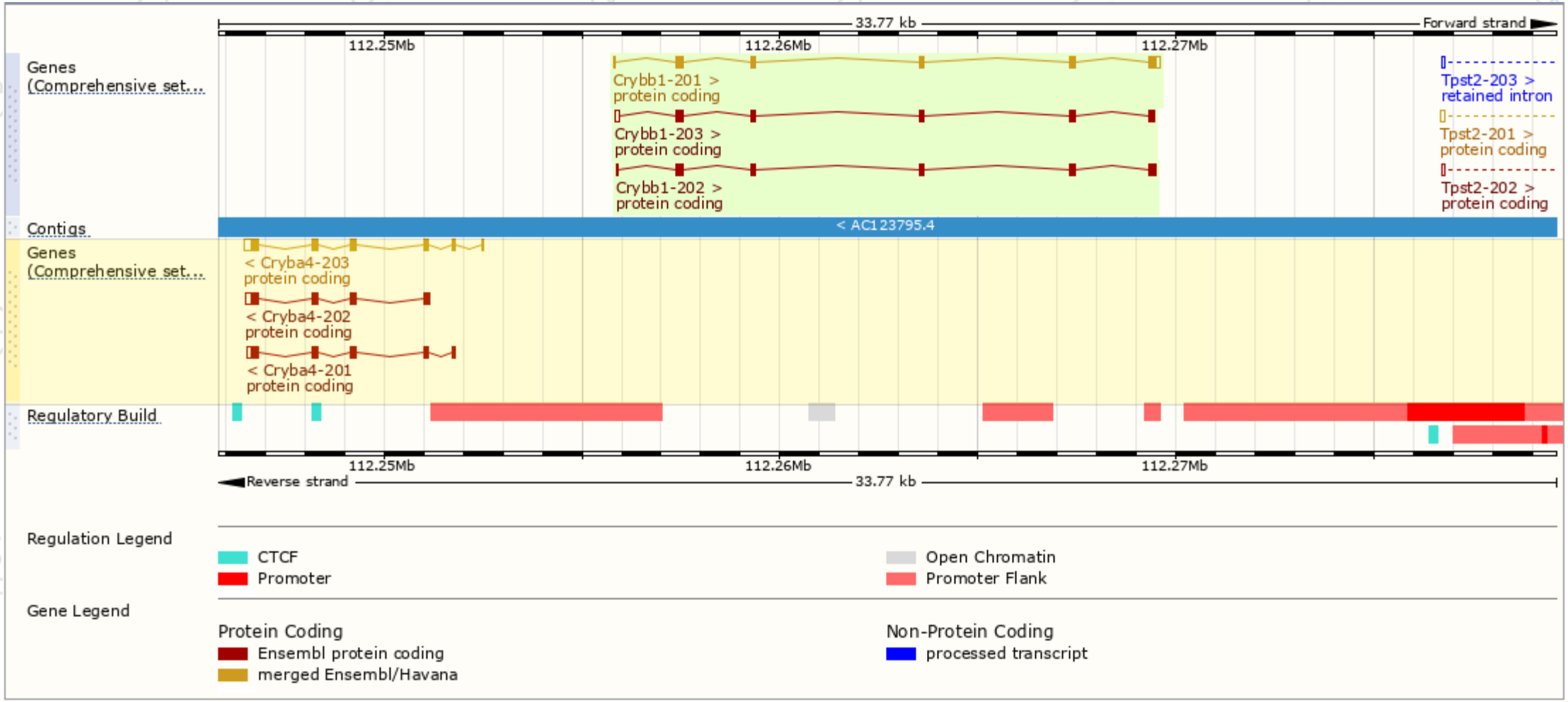
The gene has 3 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Crybb1-201	ENSMUST00000031286.12	882	250aa	Protein coding	CCDS19537	Q9WVJ5	TSL:1 GENCODE basic APPRIS P1
Crybb1-202	ENSMUST00000112375.1	793	250aa	Protein coding	CCDS19537	Q9WVJ5	TSL:3 GENCODE basic APPRIS P1
Crybb1-203	ENSMUST00000131673.7	827	236aa	Protein coding	-	E9PYP8	CDS 3' incomplete TSL:2

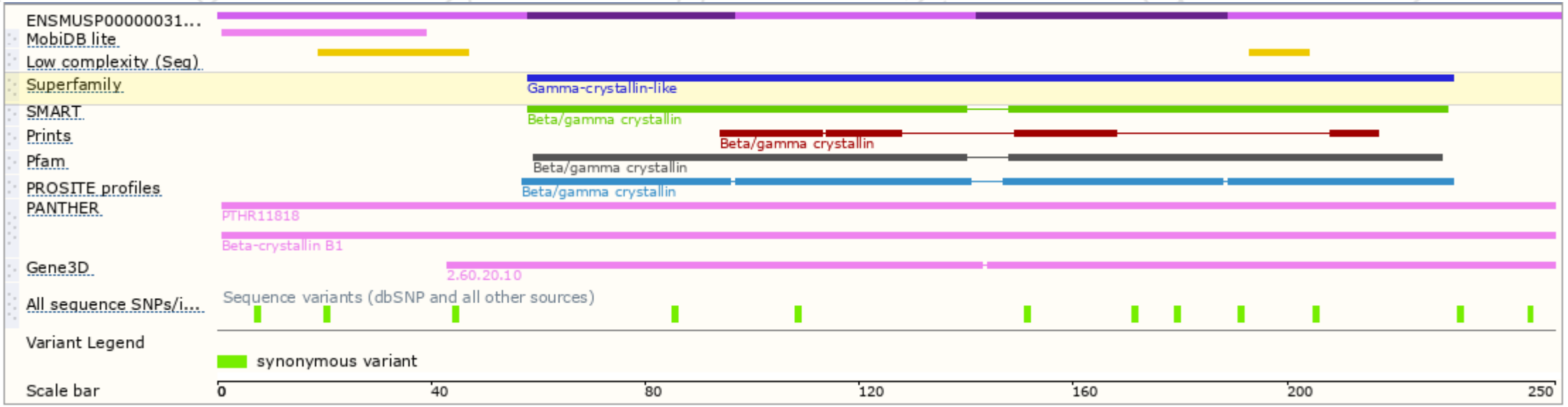
The strategy is based on the design of *Crybb1*-201 transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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