Crtap Cas9-CKO *Strategy*

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Project Overview

Project Name	***Crtap***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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

Technical routes

- The *Crtap* gene has 2 transcript.According to the structure of *Crtap* gene, exon1-3 of *Crtap*-201 transcript is recommended as the knockout region.The region contains start codon ATG coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Crtap* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 or cell types.

Notice

- According to the existing MGI data　Homozygotes develop kyphoscoliosis and osteoporosis as a result of defects in bone formation.

- The *Crtap* gene is located on the Chr 9. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information NCBI

集萃药康 GemPharmatech

Crtap cartilage associated protein [*Mus musculus* (house mouse)]

Gene ID: 56693, updated on 31-Jan-2019

Summary

Official Symbol	Crtap provided by MGI
Official Full Name	cartilage associated protein provided by MGI
Primary source	MGI:MGI:1891221
See related	Ensembl:ENSMUSG00000032431
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	CASP; P3h5; Leprel3; 5730529N23Rik
Expression	Broad expression in adrenal adult (RPKM 194.8), ovary adult (RPKM 168.2) and 16 other tissues See more
Orthologs	human all

Transcript information Ensembl

The gene has 2 transcripts, and all transcripts are shown below:

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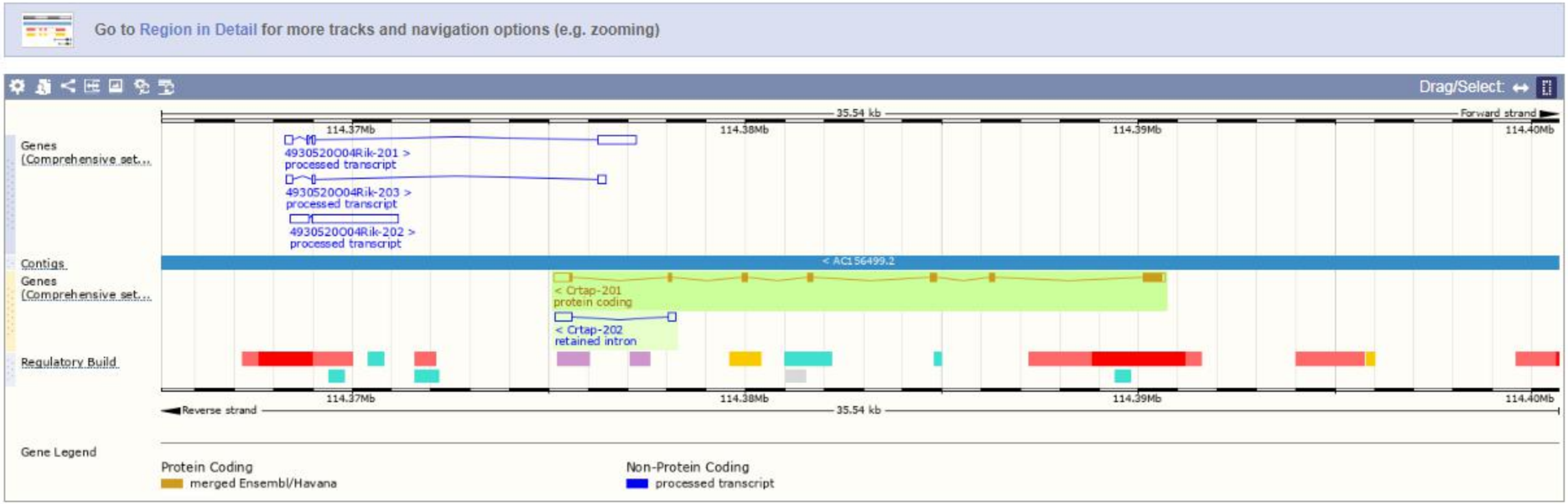
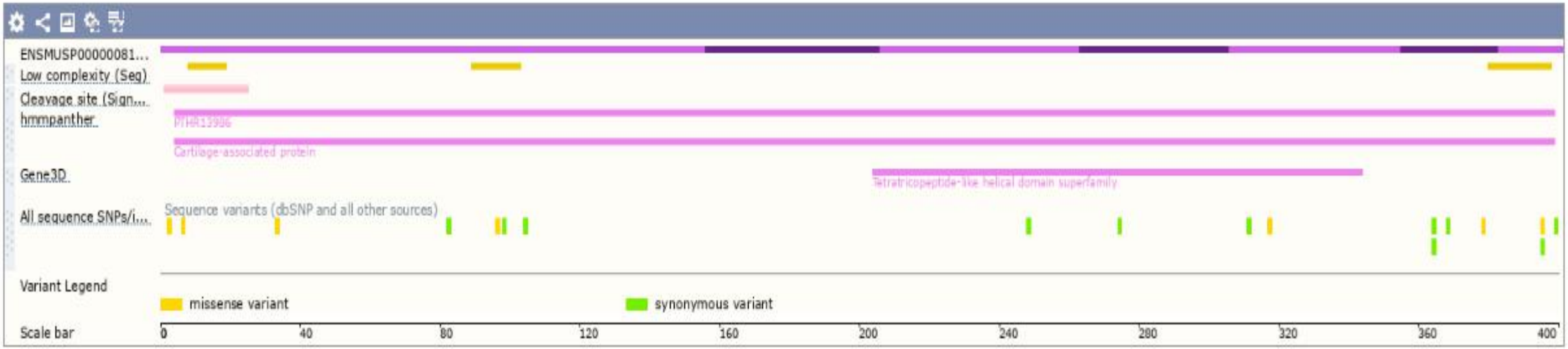
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Crtap-201	ENSMUST00000084881.4	1674	400aa	Protein coding	CCDS23592	Q9CYD3	TSL:1 GENCODE basic APPRIS P1
Crtap-202	ENSMUST00000151377.1	615	No protein	Retained intron	-	-	TSL:2

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

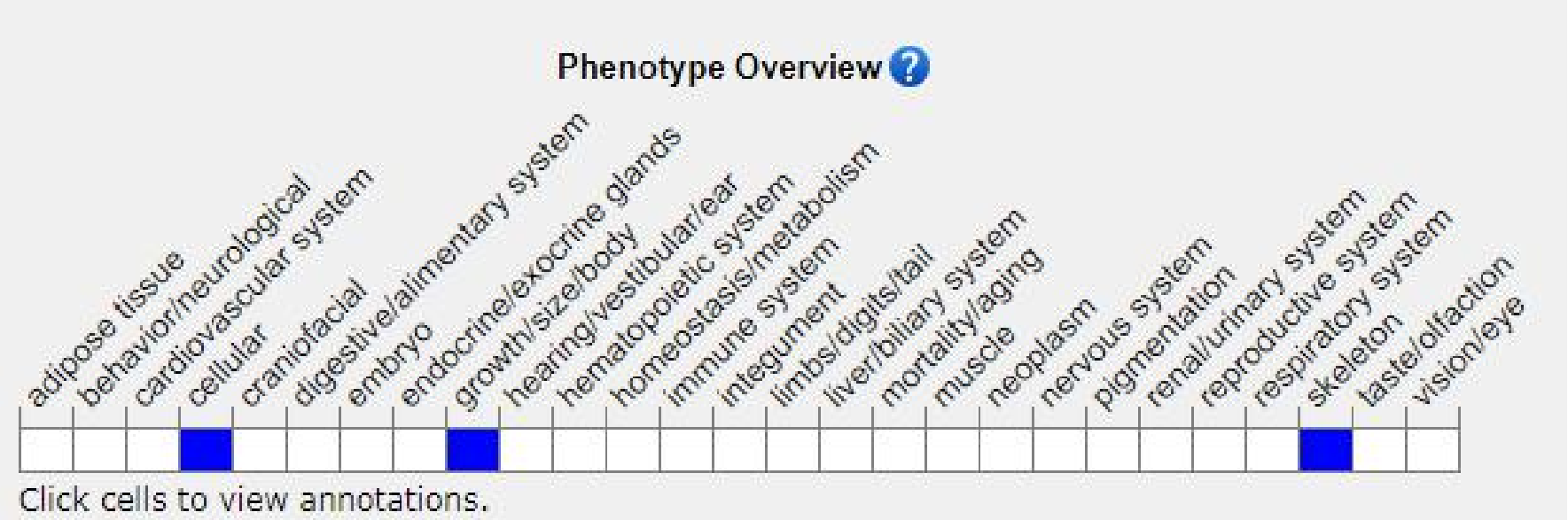
Genomic location distribution



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/) .

According to the existing MGI data,Mutations in this locus affect cell-cycle regulation and apoptos is. Null homozygotes show high, early-onset tumor incidence; some have persistent hyaloid vasculature and cataracts. Truncated or temperature-sensitive alleles cause early aging phenotypes.

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