

***Cabp2* Cas9-KO Strategy**

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Design Date: 2020-8-24

Project Overview

Project Name

Cabp2

Project type

Cas9-KO

Strain background

C57BL/6JGpt

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cabp2* gene. The schematic diagram is as follows:



- The *Cabp2* gene has 5 transcripts. According to the structure of *Cabp2* gene, exon3-exon7 of *Cabp2*-205(ENSMUST00000162908.1) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cabp2* gene. The brief process is as follows: sgRNA was transcribed in vitro. Cas9 and sgRNA 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.

- According to the existing MGI data, homozygous knockout affects calcium channels in cochlear inner hair cell synapses, resulting in hearing impairment. It also affects transmission of responses to light through the retinal circuits.
- The *Cabp2* gene is located on the Chr19. 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)

Cabp2 calcium binding protein 2 [Mus musculus (house mouse)]

Gene ID: 29866, updated on 13-Mar-2020

Summary

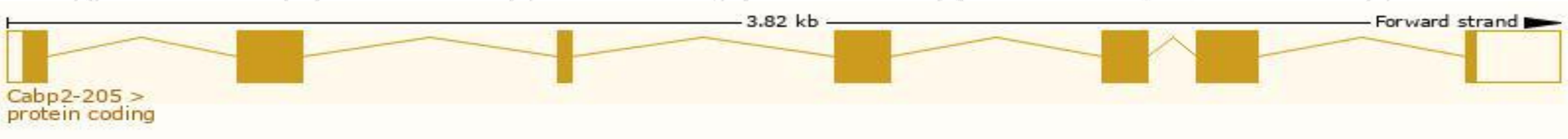
Official Symbol	Cabp2 provided by MGI
Official Full Name	calcium binding protein 2 provided by MGI
Primary source	MGI:MGI:1352749
See related	Ensembl:ENSMUSG00000024857
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Biased expression in small intestine adult (RPKM 1.9), spleen adult (RPKM 1.5) and 4 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

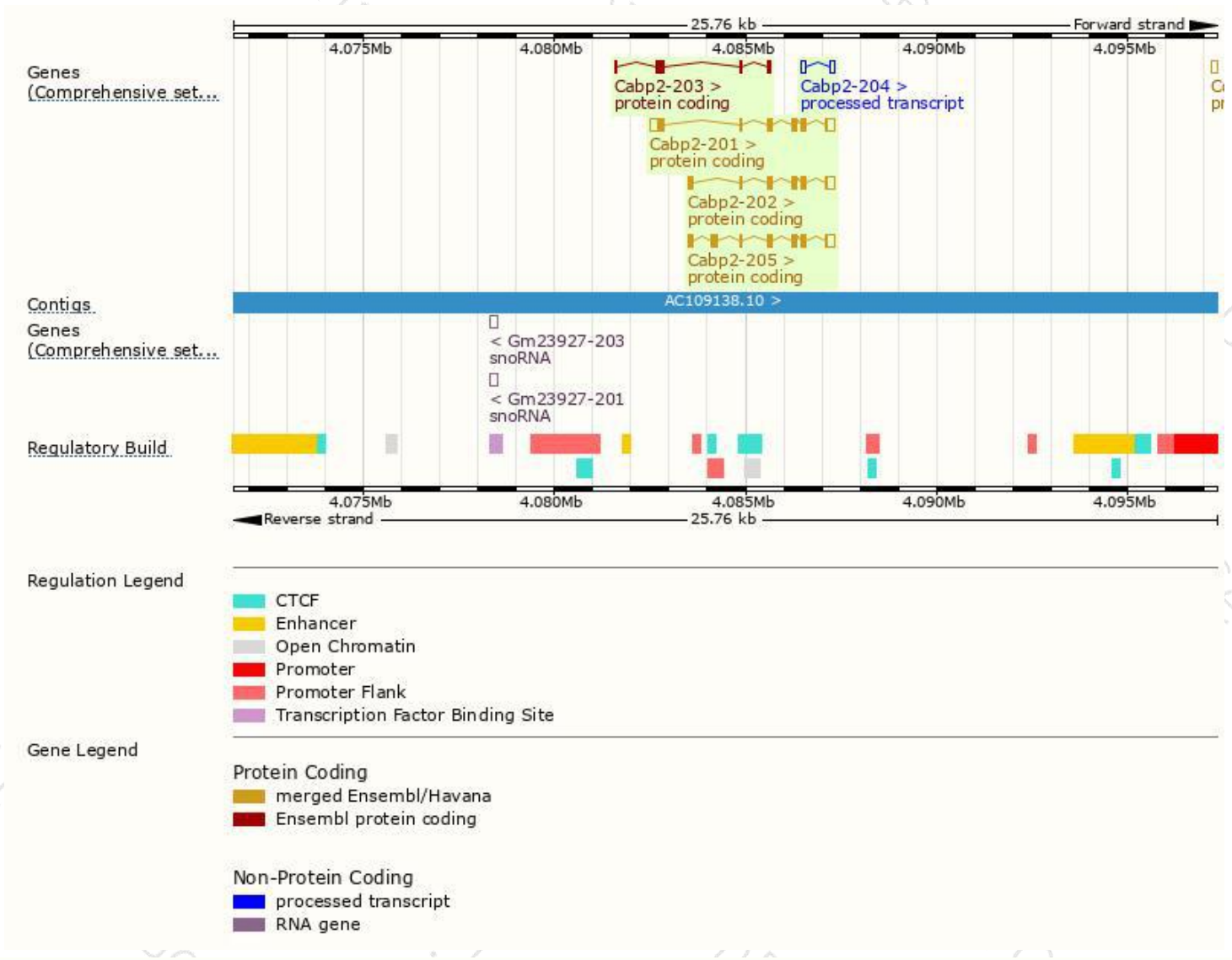
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cabp2-201	ENSMUST00000159148.7	1030	203aa	Protein coding	CCDS50348	G5E8W3	TSL:1 GENCODE basic APPRIS ALT2
Cabp2-205	ENSMUST00000162908.1	911	221aa	Protein coding	CCDS29413	G3XA29	TSL:1 GENCODE basic APPRIS P3
Cabp2-202	ENSMUST00000159556.7	781	168aa	Protein coding	CCDS50349	G3XA16	TSL:1 GENCODE basic APPRIS ALT2
Cabp2-203	ENSMUST00000159593.7	387	103aa	Protein coding	-	E0CXZ2	CDS 3' incomplete TSL:5
Cabp2-204	ENSMUST00000162584.1	243	No protein	Processed transcript	-	-	TSL:3

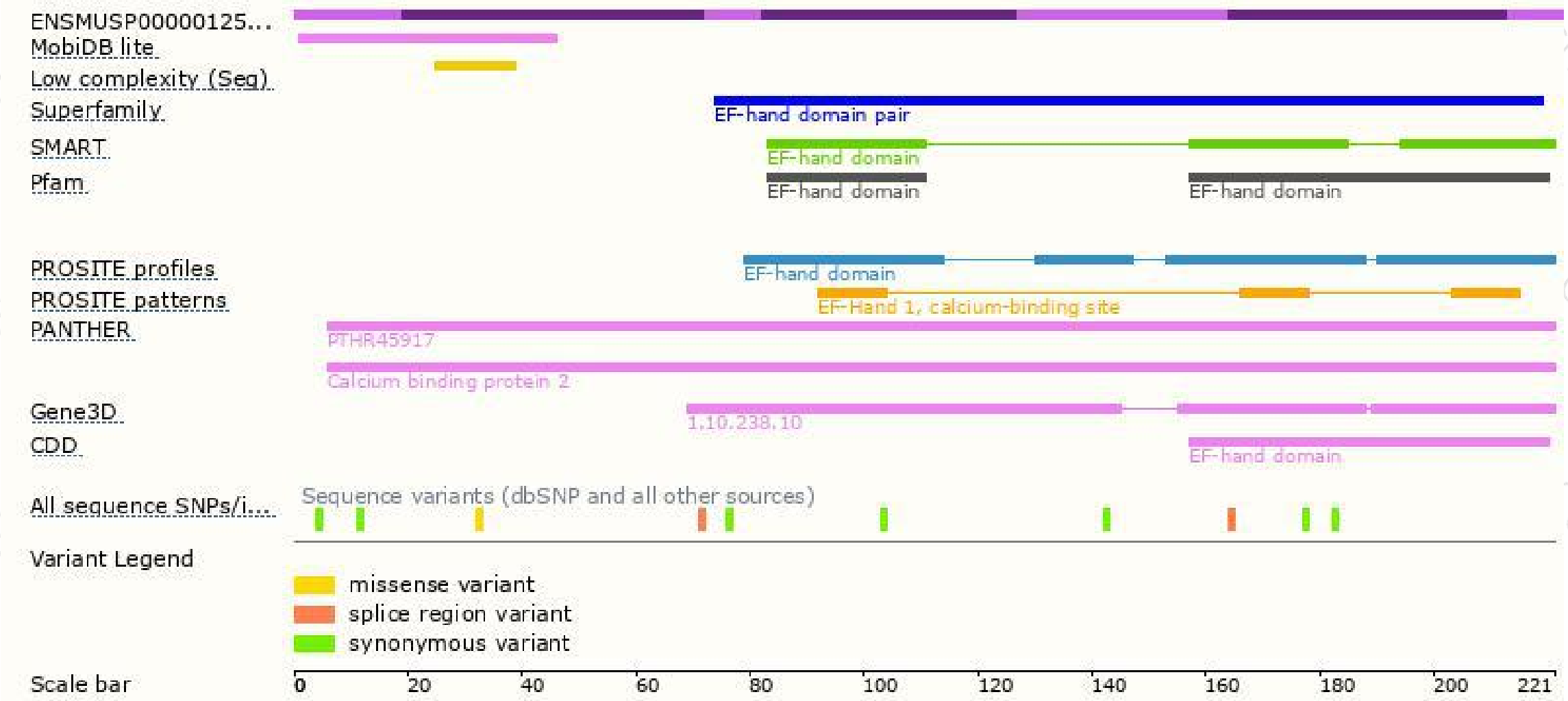
The strategy is based on the design of *Cabp2-205* transcript,the transcription is shown below:



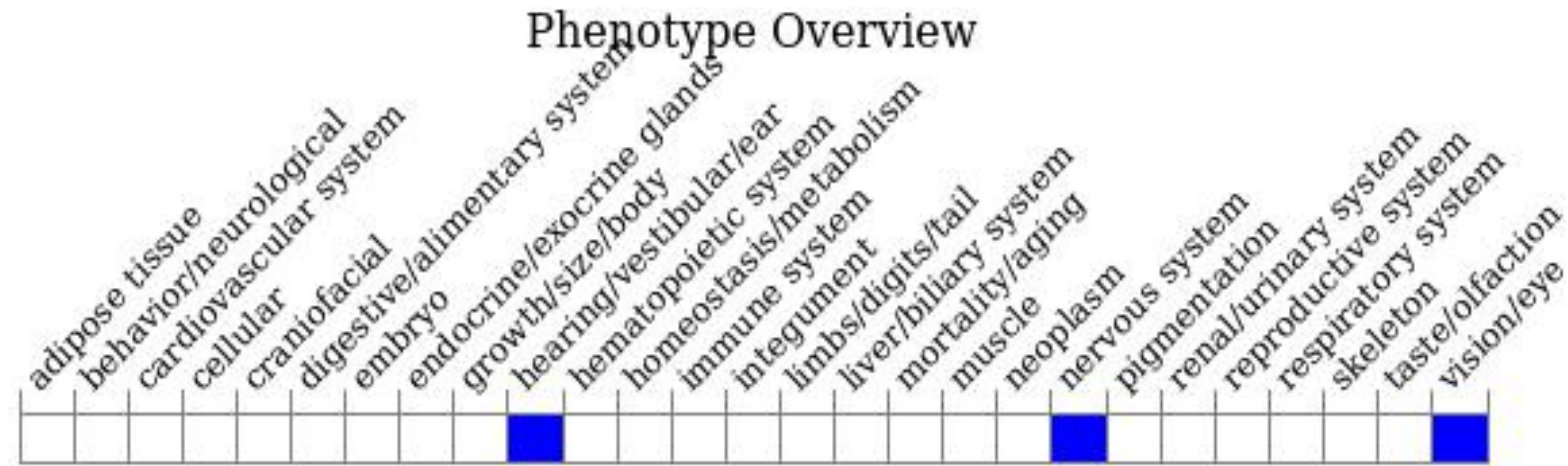
Genomic location distribution



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, homozygous knockout affects calcium channels in cochlear inner hair cell synapses, resulting in hearing impairment. It also affects transmission of responses to light through the retinal circuits.

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

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