Ephx3 Cas9-CKO Strategy

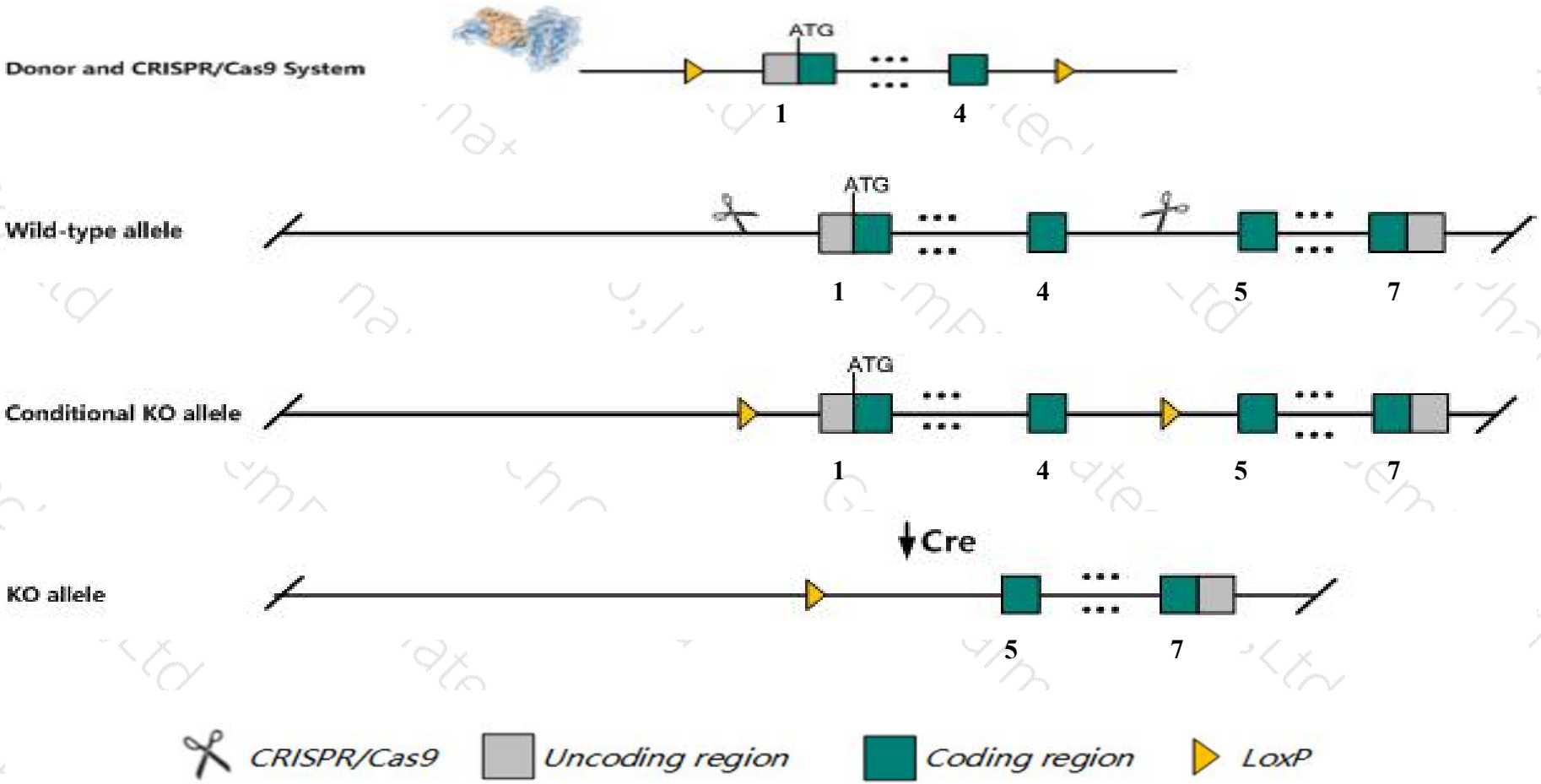
Designer:

Project Overview

Project Name	***Ephx3***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ephx3* gene. The schematic diagram is as follows:



Technical routes

- The *Ephx3* gene has 2 transcripts. According to the structure of *Ephx3* gene, exon1-exon4 of *Ephx3-201* (ENSMUST00000087721.9) transcript is recommended as the knockout region.The region contains start codon ATG. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Ephx3* 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 and cell types.

Notice

- According to the existing MGI data,Mice homozygous for a null allele are viable and fertile with no gross abnormalities.

- The *Ephx3* gene is located on the Chr17. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information （NCBI）

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Ephx3 epoxide hydrolase 3 [Mus musculus (house mouse)]

Gene ID: 71932, updated on 31-Jan-2019

Summary

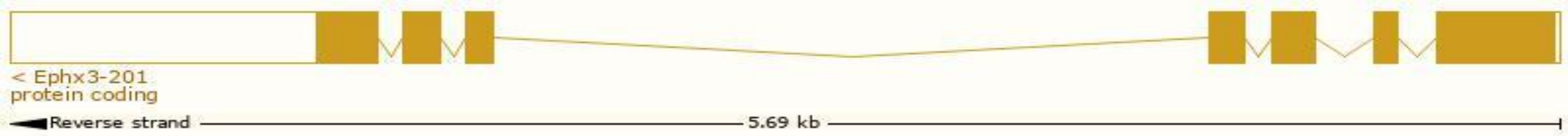
Official Symbol	Ephx3 provided by MGI
Official Full Name	epoxide hydrolase 3 provided by MGI
Primary source	MGI:MGI:1919182
See related	Ensembl:ENSMUSG00000037577
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2310063B19Rik, Abhd9, EH3
Expression	Biased expression in stomach adult (RPKM 8.4), lung adult (RPKM 2.9) and 6 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

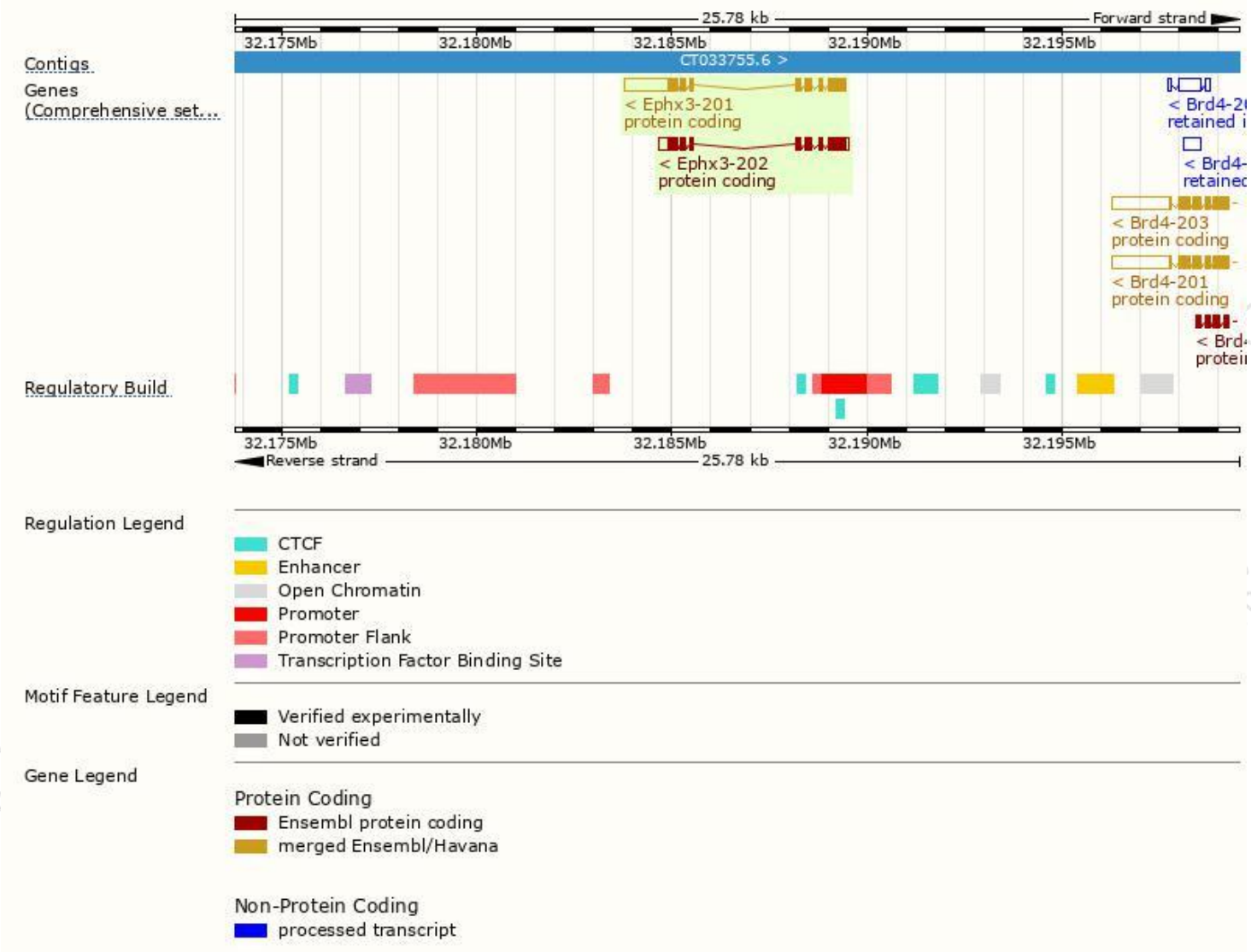
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ephx3-201	ENSMUST00000087721.9	2416	424aa	Protein coding	CCDS28615	A0A0R4J127	TSL:1 GENCODE basic APPRIS P3
Ephx3-202	ENSMUST00000162117.2	1591	417aa	Protein coding	CCDS84290	G3XA19	TSL:1 GENCODE basic APPRIS ALT2

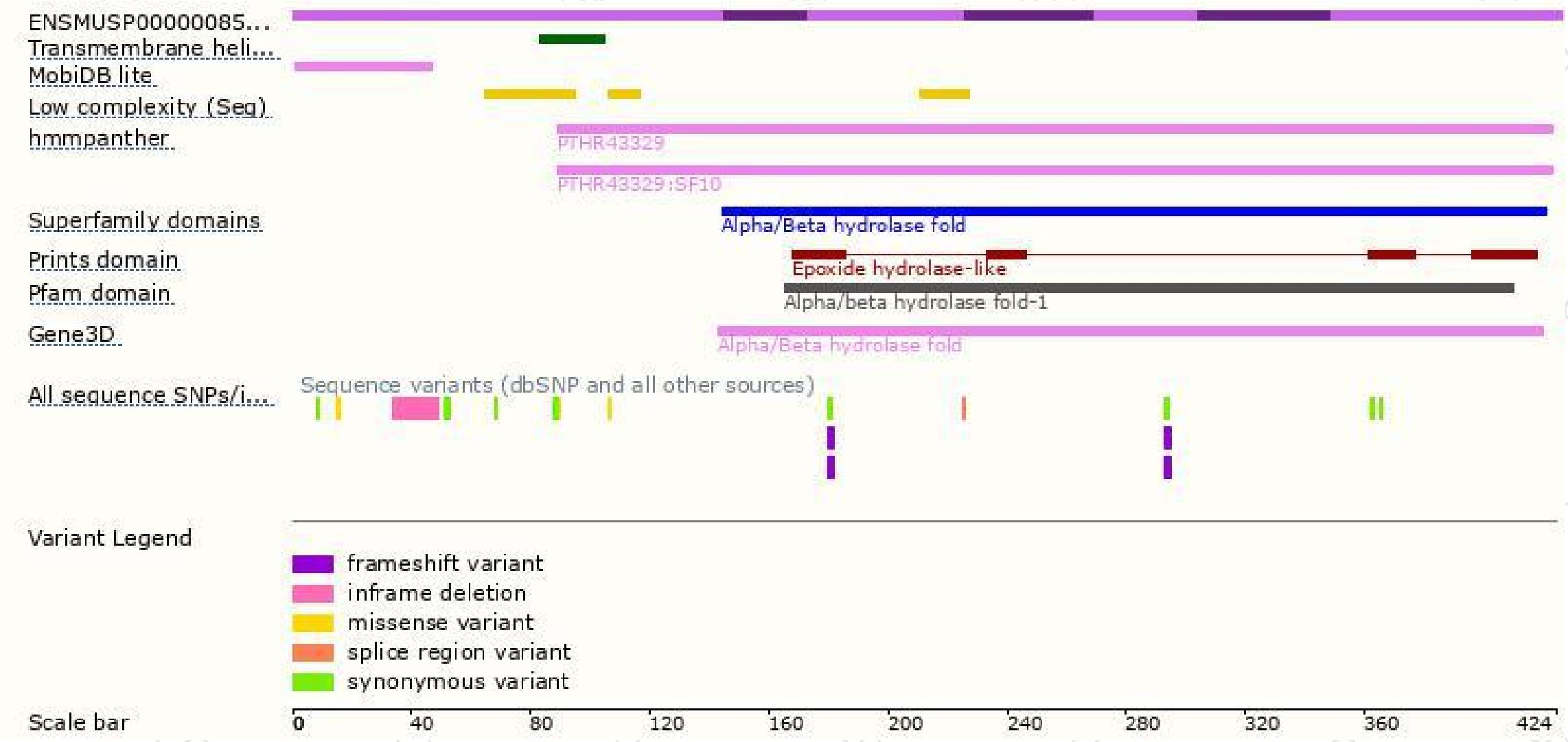
The strategy is based on the design of *Ephx3-201* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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