

Cpa3 Cas9-CKO Strategy

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Project Overview

Project Name

Cpa3

Project type

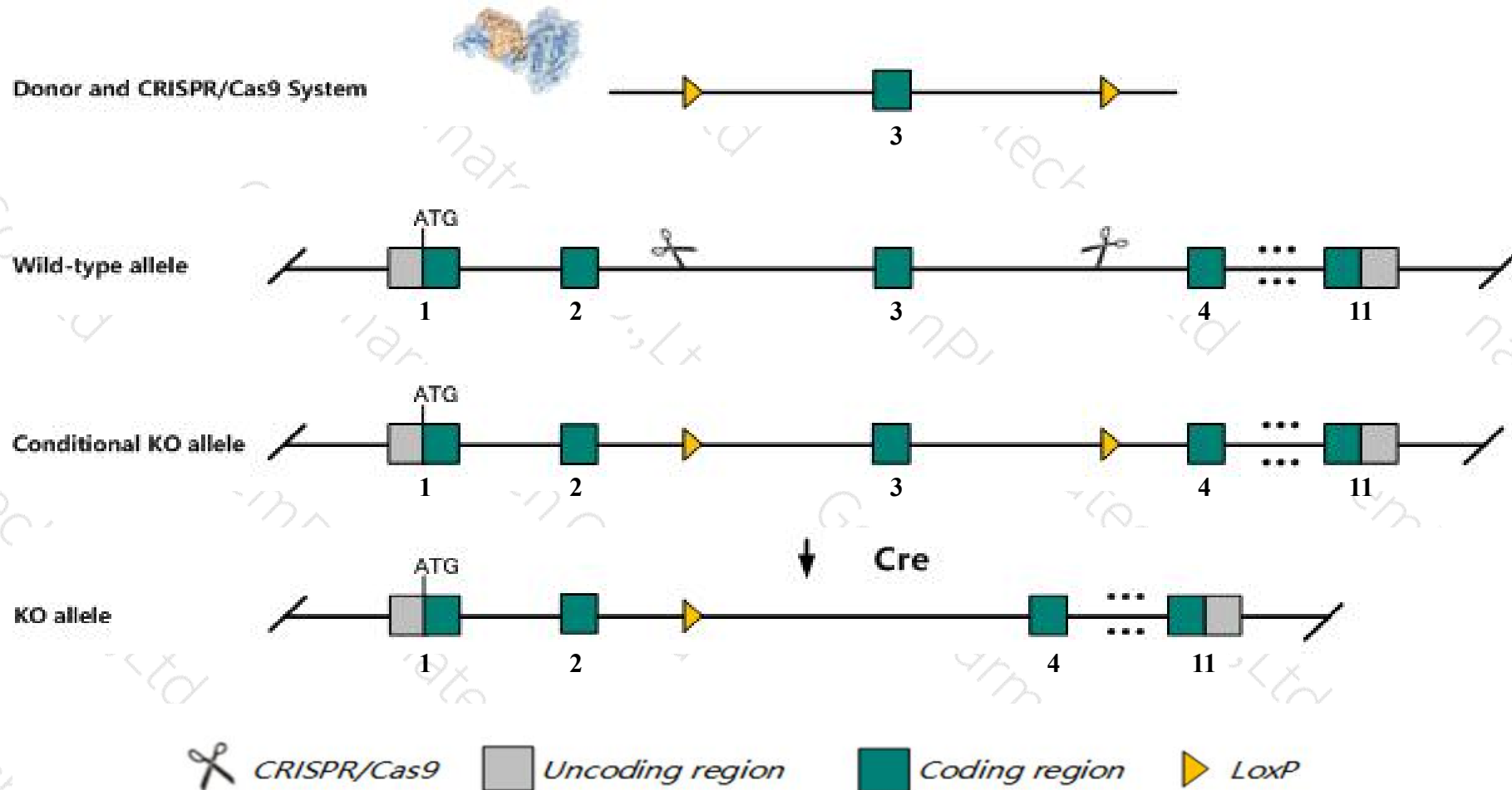
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cpa3* gene. The schematic diagram is as follows:



Technical routes

- The *Cpa3* gene has 2 transcripts. According to the structure of *Cpa3* gene, exon3 of *Cpa3-201* (ENSMUST00000001921.2) transcript is recommended as the knockout region. The region contains 125bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cpa3* gene. The brief process is as follows: CRISPR/Cas9 system 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.

- According to the existing MGI data, Homozygous null mice have immature peritoneal mast cells but normal mast cell functions.
- The *Cpa3* gene is located on the Chr3. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Cpa3 carboxypeptidase A3, mast cell [*Mus musculus* (house mouse)]

Gene ID: 12873, updated on 22-Oct-2019

Summary

Official Symbol	Cpa3 provided by MGI
Official Full Name	carboxypeptidase A3, mast cell provided by MGI
Primary source	MGI:MGI:88479
See related	Ensembl:ENSMUSG00000001865
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	MC-CPA
Summary	This gene encodes a member of the carboxypeptidase A family of zinc metalloproteases and preproprotein that is proteolytically processed to generate a mature protein product. This product is released by mast cells and may be involved in the degradation of endogenous proteins and the inactivation of venom-associated peptides. Homozygous knockout mice for this gene exhibit impaired mast cell development. [provided by RefSeq, Aug 2015]
Expression	Biased expression in limb E14.5 (RPKM 23.7), mammary gland adult (RPKM 5.5) and 4 other tissues See more
Orthologs	human all

Genomic context

Location: 3 A2; 3 6.25 cM

Exon count: 11

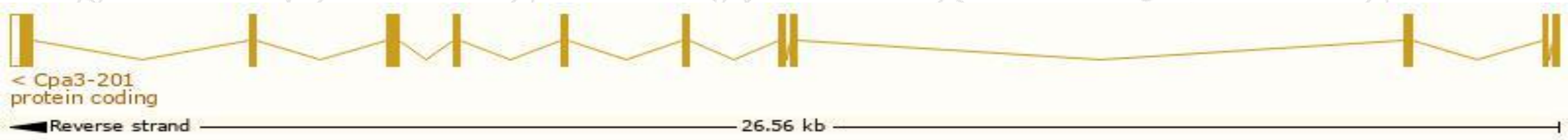
See Cpa3 in [Genome Data Viewer](#)

Transcript information (Ensembl)

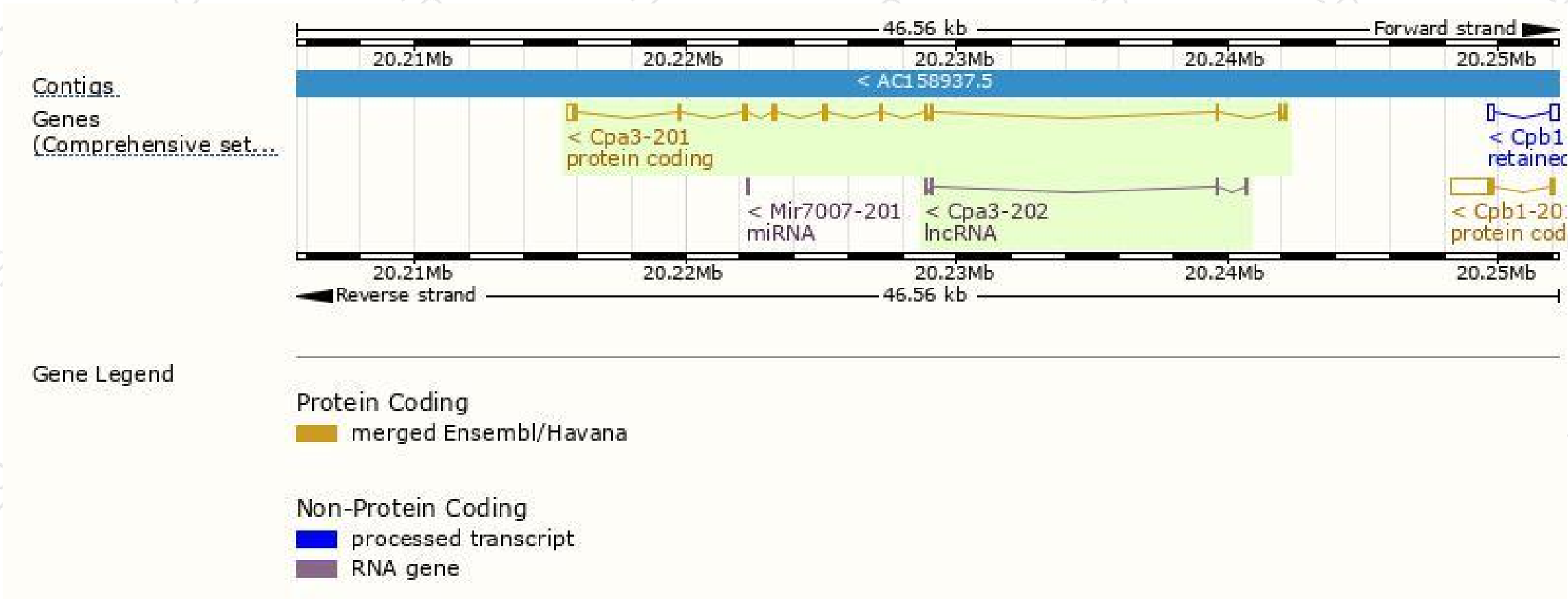
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cpa3-201	ENSMUST00000001921.2	1449	417aa	Protein coding	CCDS17262	P15089 Q542E3	TSL:1 GENCODE basic APPRIS P1
Cpa3-202	ENSMUST00000191659.1	398	No protein	lncRNA	-	-	TSL:3

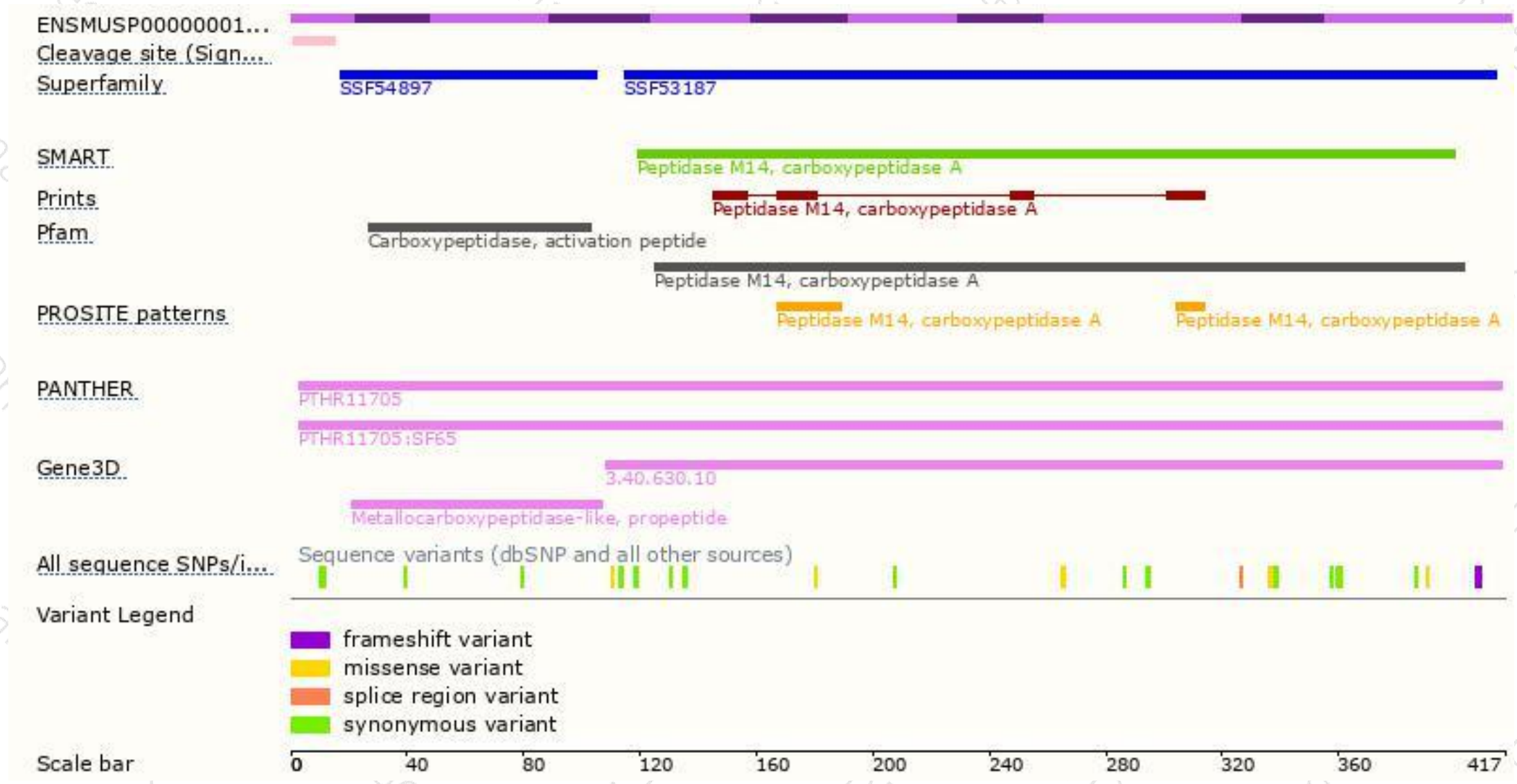
The strategy is based on the design of *Cpa3-201* transcript,The transcription is shown below



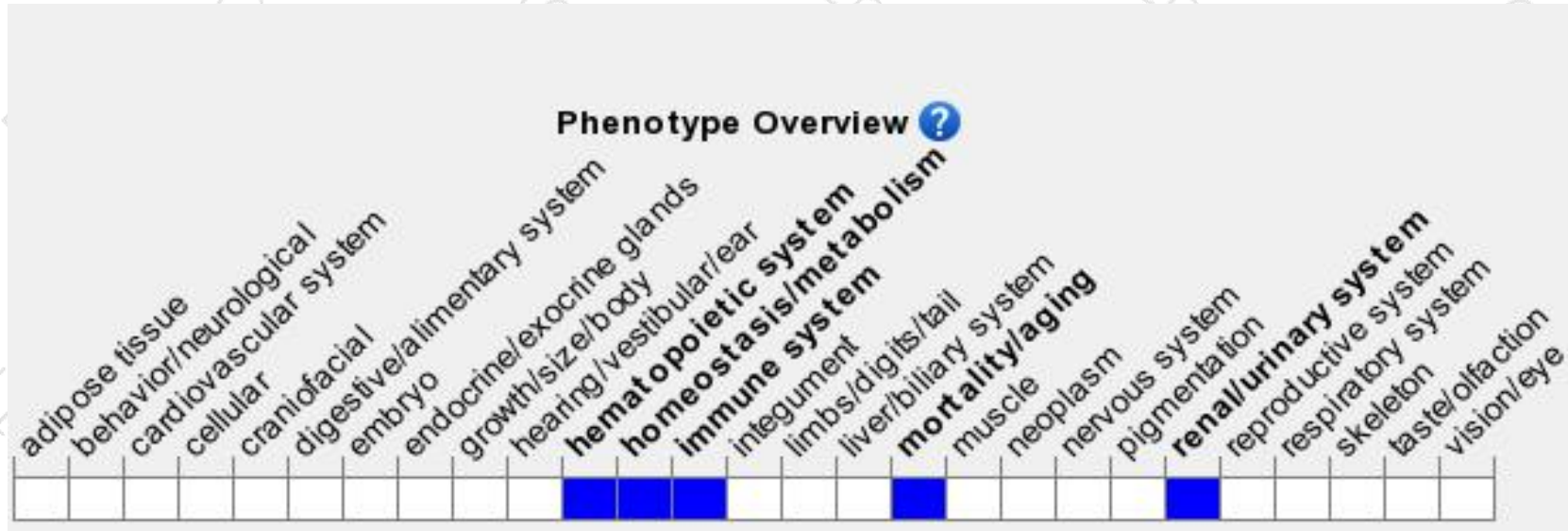
Genomic location distribution



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, Homozygous null mice have immature peritoneal mast cells but normal mast cell functions.

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

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