

***Pax7-DreERT2-P2A* Cas9-KI Mouse Model Strategy**

Designer: Hui Li

Reviewer: Daohua Xu

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Project Overview

Project Name

Pax7-DreERT2-P2A

Project type

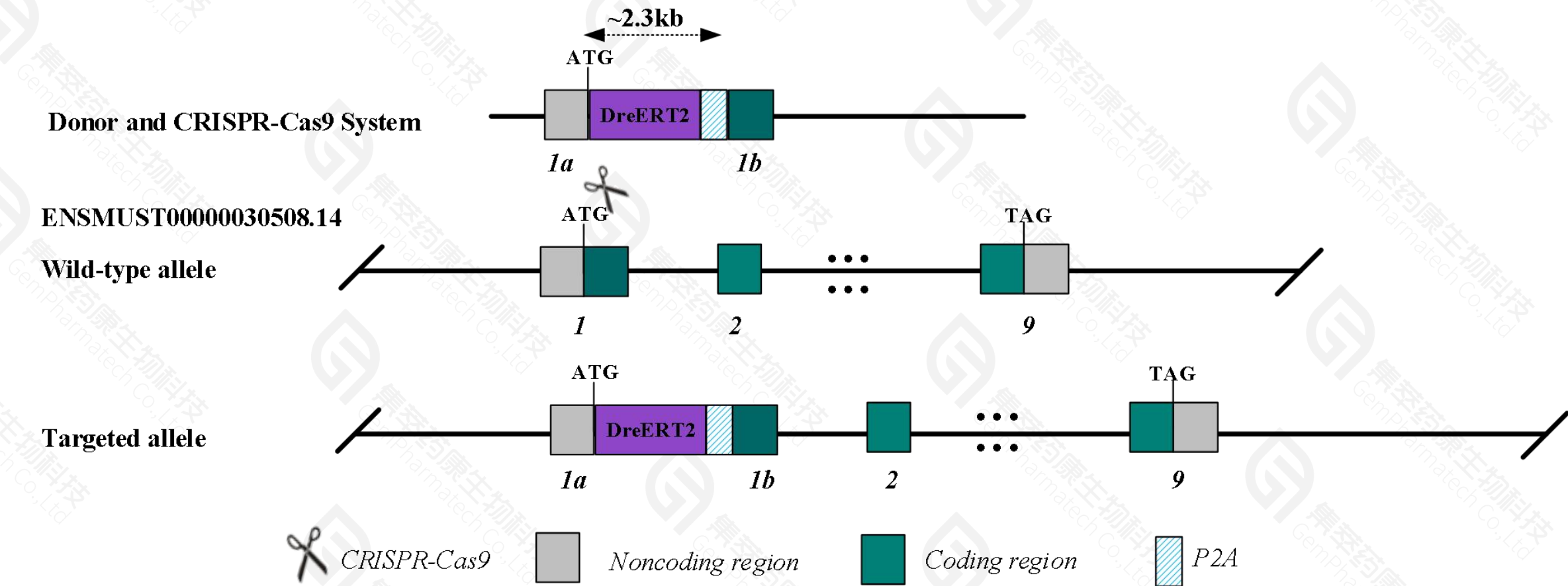
Cas9-KI

Strain background

C57BL/6JGpt

Knock in strategy

This model will use CRISPR-Cas9 technology to edit the *Pax7* gene. The schematic diagram is as follows:



Technical routes

- The *Pax7* gene has 2 transcripts.
- According to the structure of *Pax7* gene and customer request, the element *DreERT2-P2A* will be inserted at the translation start codon of *Pax7* -201(ENSMUST00000030508.14), the length of inserted fragment is about 2.3 kb.
- The mouse *Pax7* -201 transcript contains 9 exons. The translation initiation site ATG is located at exon 1, and the translation termination site TAG is located at exon 9, encoding 503 aa.
- In this project we use CRISPR/Cas9 technology to modify *Pax7* 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.

- According to the existing MGI data, Mice homozygous for a targeted null mutation exhibit craniofacial malformations involving the nose and maxilla, and die within three weeks after birth.
- One or two synonymous mutations of amino acids may be introduced on exon1
- The upstream and downstream sequence of knockin site contained multiple repeat structures, mutations may be introduced during the model production.
- The P2A-linked gene drives expression in the same promoter and is cleaved at the translational level. The gene expression levels are consistent, and the before of P2A expressing gene carries the P2A-translated polypeptide.
- The *Pax7* gene is located on the Chr4. Please take the loci in consideration when breeding this knockin mice with other gene modified strains, if the other gene is also on Chr4, it may be extremely hard to get double gene positive homozygotes.
- The scheme is designed according to the genetic information in the existing database. Inserting a foreign gene between the 5'UTR and the gene coding region may affect the expression of endogenous and foreign genes. Due to the complexity of biological processes, it cannot be predicted completely at the present technology level.

Gene information (NCBI)

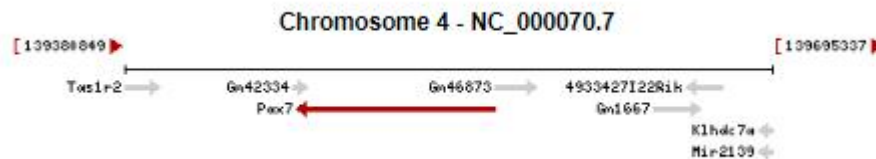
Pax7 paired box 7 [*Mus musculus* (house mouse)]

[Download Datasets](#)

Gene ID: 18509, updated on 28-Jun-2022

Summary

Official Symbol	Pax7 provided by MGI
Official Full Name	paired box 7 provided by MGI
Primary source	MGI:MGI:97491
See related	Ensembl:ENSMUSG00000028736 AllianceGenome:MGI:97491
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<i>Mus musculus</i>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Pax-7
Summary	Enables DNA-binding transcription factor activity, RNA polymerase II-specific and sequence-specific DNA binding activity. Acts upstream of or within several processes, including animal organ development; chordate embryonic development; and positive regulation of nitrogen compound metabolic process. Located in nucleus. Is expressed in several structures, including central nervous system; embryo mesenchyme; genitourinary system; sensory organ; and skeletal musculature. Human ortholog(s) of this gene implicated in alveolar rhabdomyosarcoma. Orthologous to human PAX7 (paired box 7). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Biased expression in whole brain E14.5 (RPKM 2.5), CNS E11.5 (RPKM 2.3) and 4 other tissues See more
Orthologs	human all
NEW	Try the new Gene table Try the new Transcript table



Transcript information (Ensembl)

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

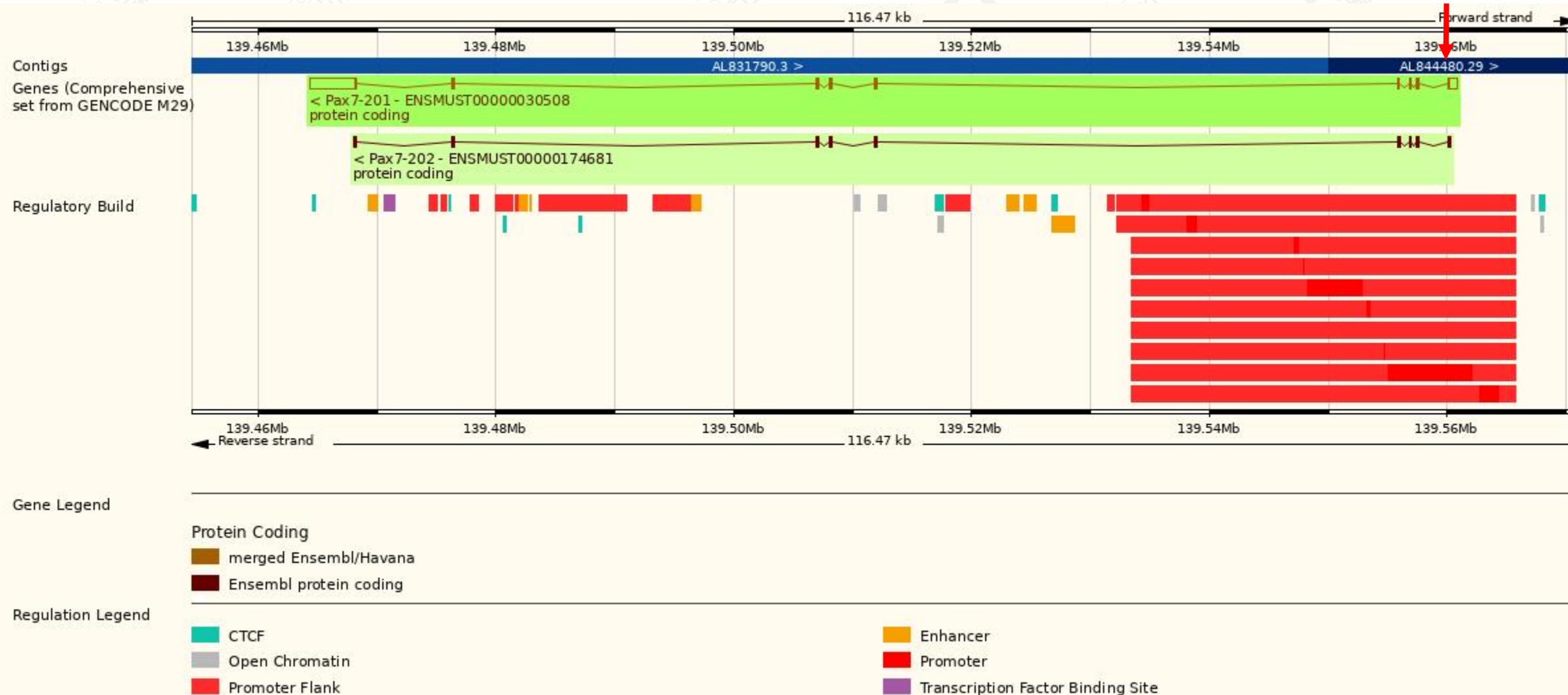
Show/hide columns (1 hidden)		Filter					
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
ENSMUST00000030508.14	Pax7-201	5854	503aa	Protein coding	CCDS18850	P47239	GENCODE basic APPRIS P1 TSL:1
ENSMUST00000174681.2	Pax7-202	1725	505aa	Protein coding		G3UX36	Ensembl Canonical GENCODE basic TSL:5

The strategy is based on the design of *Pax7* -201 transcript, the transcription is shown below:

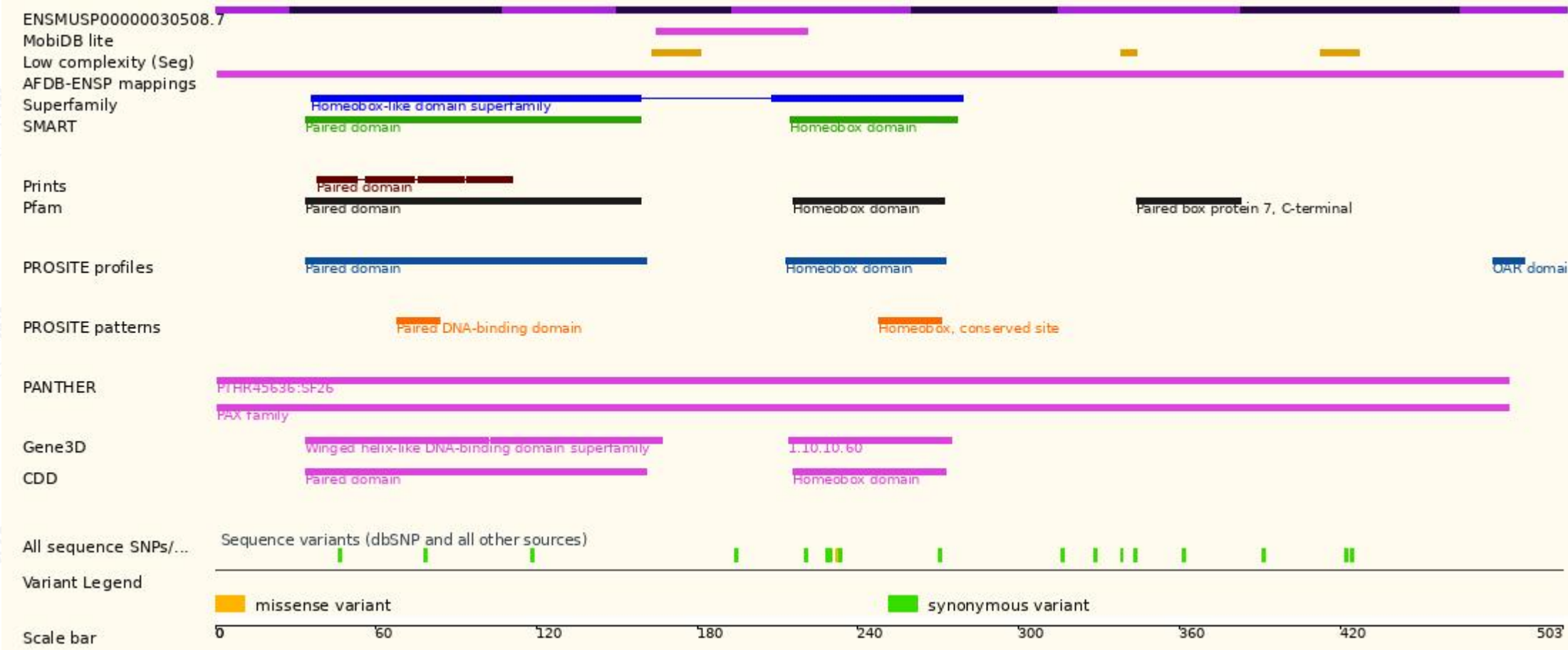


Genomic location distribution

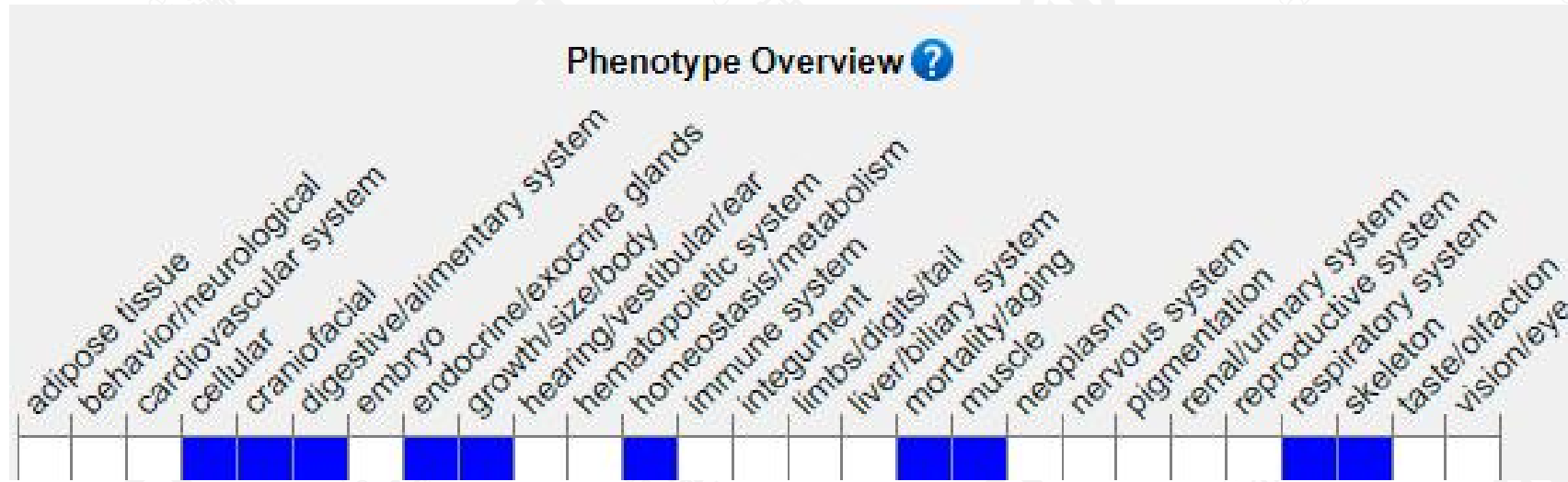
insertion site



Protein domains for ENSMUSP00000030508.7



Mouse phenotype description(MGI)



Mice homozygous for a targeted null mutation exhibit craniofacial malformations involving the nose and maxilla, and die within three weeks after birth. Mice homozygous for floxed alleles activated in muscle cells exhibit reduced satellite cell numbers and impaired muscle regeneration.

<http://www.informatics.jax.org/marker/MGI:97491>

If you have any questions, you are welcome to inquire.
Tel: 400-966 0890



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