

***Srf-iCre* cas9-ki Mouse Model Strategy**

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Project Overview

Project Name

Srf-iCre

Project type

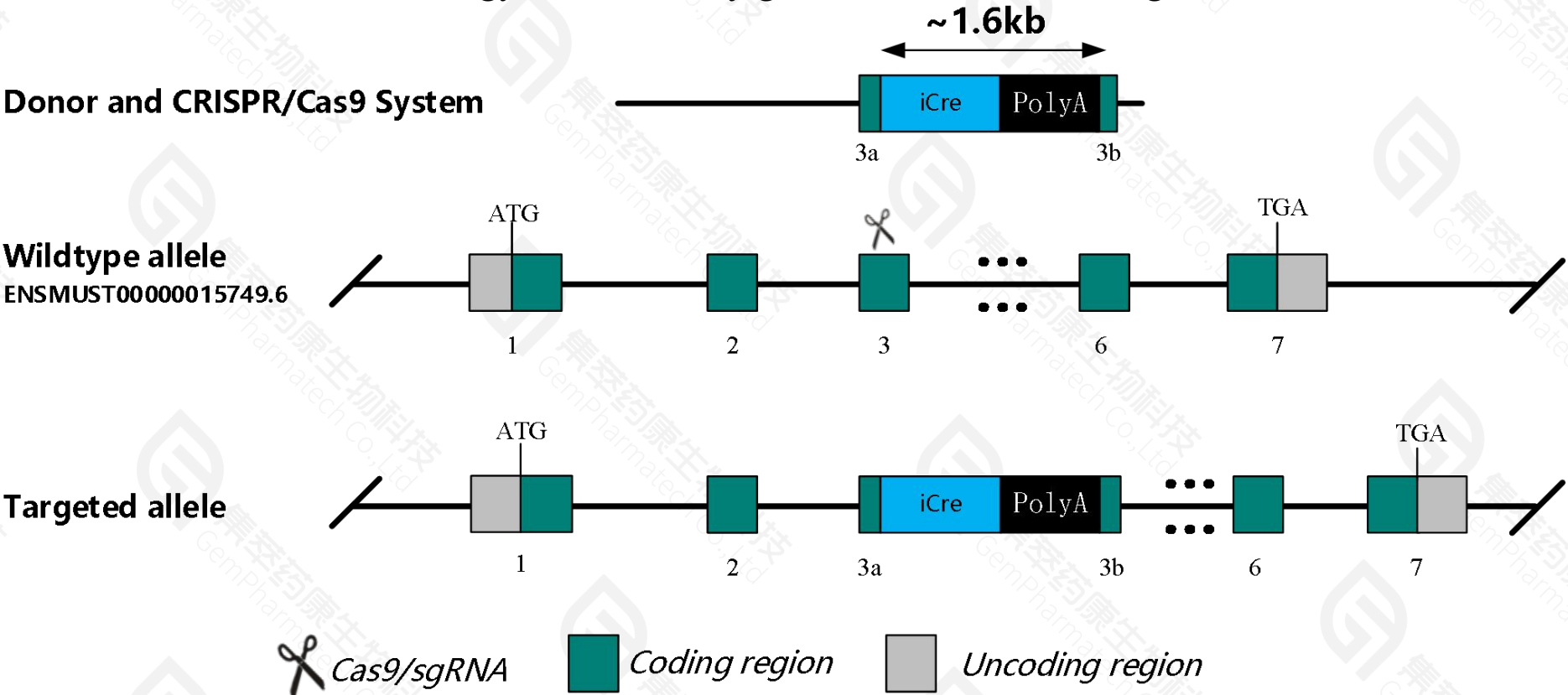
cas9-ki

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



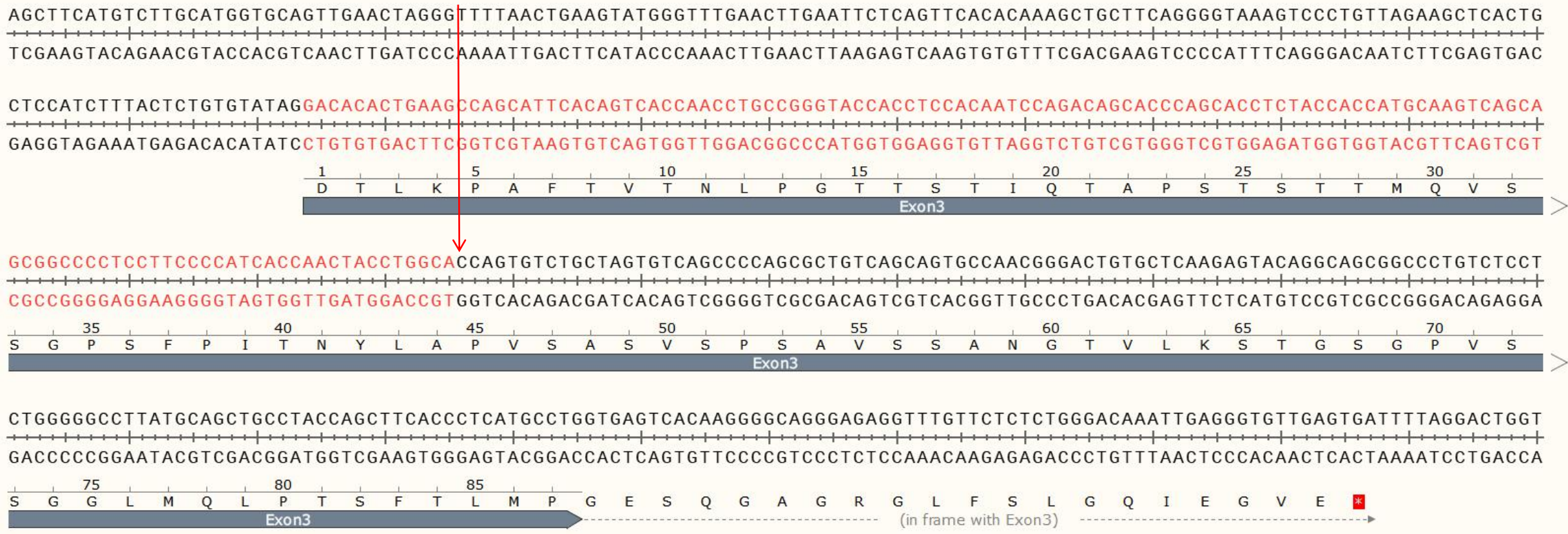
iCre will be inserted into exon3 and expressed in the frame with the amino acid at the N-terminal of the insertion site, and endogenous *Srf* gene expression will be closed.

- The *Srf* gene has 4 transcripts.
- According to the structure of *Srf* gene, the element *iCre-PolyA* will be inserted into exon3 of *Srf-201*(ENSMUST00000015749.6), the length of inserted fragment is about 1.6kb.
- The mouse *Srf-201* transcript contains 7 exons. The translation initiation site ATG is located at exon1, and the translation termination site TGA is located at exon7, encoding 504aa.
- In this project we use CRISPR/Cas9 technology to modify *Srf* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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, homozygous null mice exhibit embryonic lethality, abnormal gastrulation, no mesoderm or primitive streak formation and reduced embryo size.
- *iCre* will be inserted in exon3 as a fusion protein, the change of protein spatial structure may affect the function of *Srf* and *iCre*.
- It is necessary to introduce 1-2 synonymous mutation in exon3.
- The insertion site is close to the 3' of *Mir6976* gene, and this strategy may affect the regulation of the 3' of the *Mir6976* gene.
- Transcript *Srf*-204 will be disrupted, transcript *Srf*-203 may not be affected, and the effect of transcript *Srf*-202 is unknown.
- The *Srf* gene is located on the Chr17. Please take the loci in consideration when breeding this knockin mice with other gene modified strains, if the other gene is also on Chr17, 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 into exon 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.

Insertion site

insertion site



Gene information (NCBI)

Srf serum response factor [*Mus musculus* (house mouse)]

Gene ID: 20807, updated on 22-Dec-2020

Summary

Official Symbol Srf provided by [MGI](#)

Official Full Name serum response factor provided by [MGI](#)

Primary source [MGI:MGI:106658](#)

See related [Ensembl:ENSMUSG00000015605](#)

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 AW049942; AW240594

Expression Ubiquitous expression in ovary adult (RPKM 59.4), colon adult (RPKM 43.3) and 28 other tissues [See more](#)

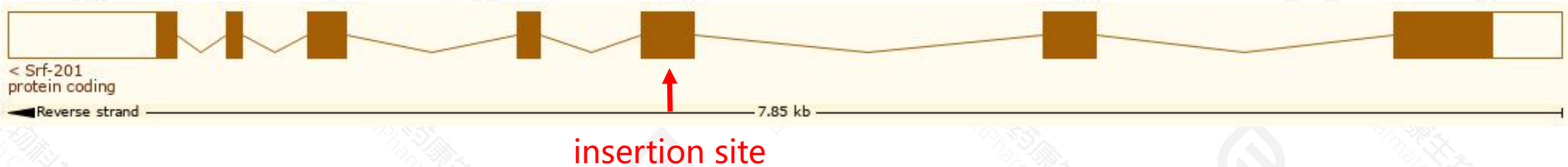
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

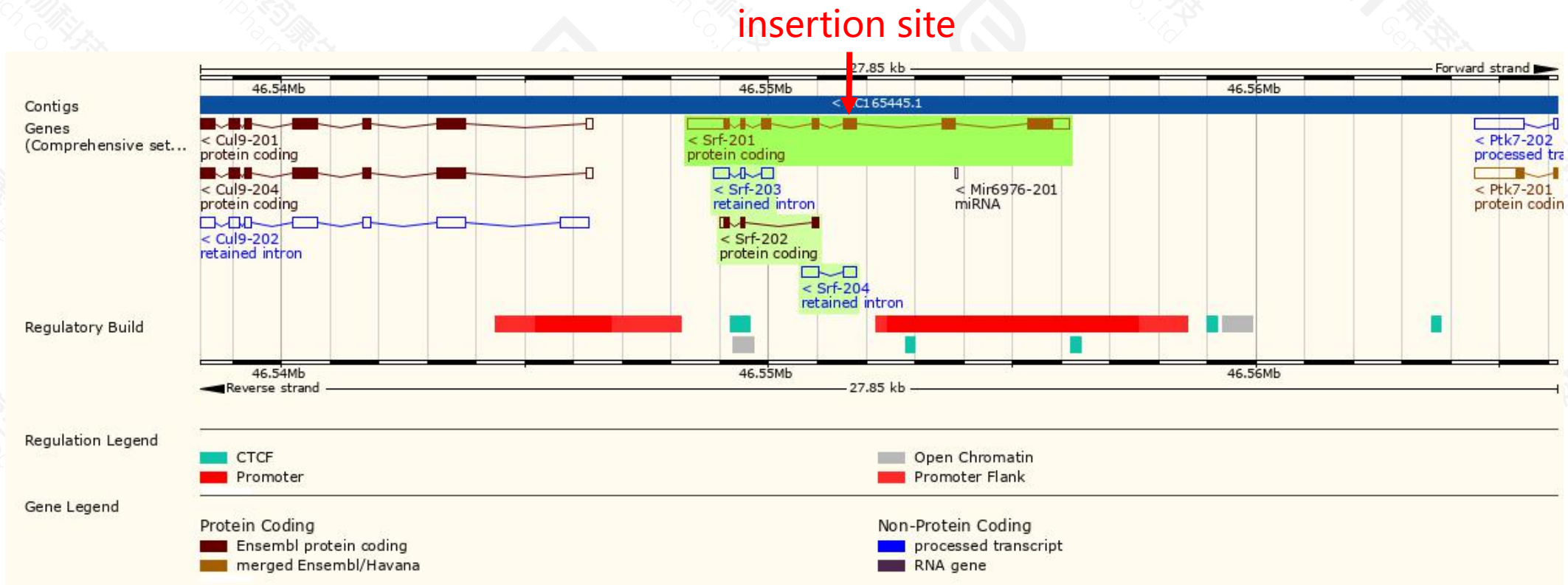
The gene has 4 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt Match	Flags
Srf-201	ENSMUST00000015749.6	2616	504aa	Protein coding	CCDS28831	Q9JM73	TSL:1 GENCODE basic APPRIS P1
Srf-202	ENSMUST00000233104.1	376	97aa	Protein coding	-	A0A3B2WCW1	CDS 5' incomplete
Srf-203	ENSMUST00000233767.1	637	No protein	Retained intron	-	-	-
Srf-204	ENSMUST00000233797.1	595	No protein	Retained intron	-	-	-

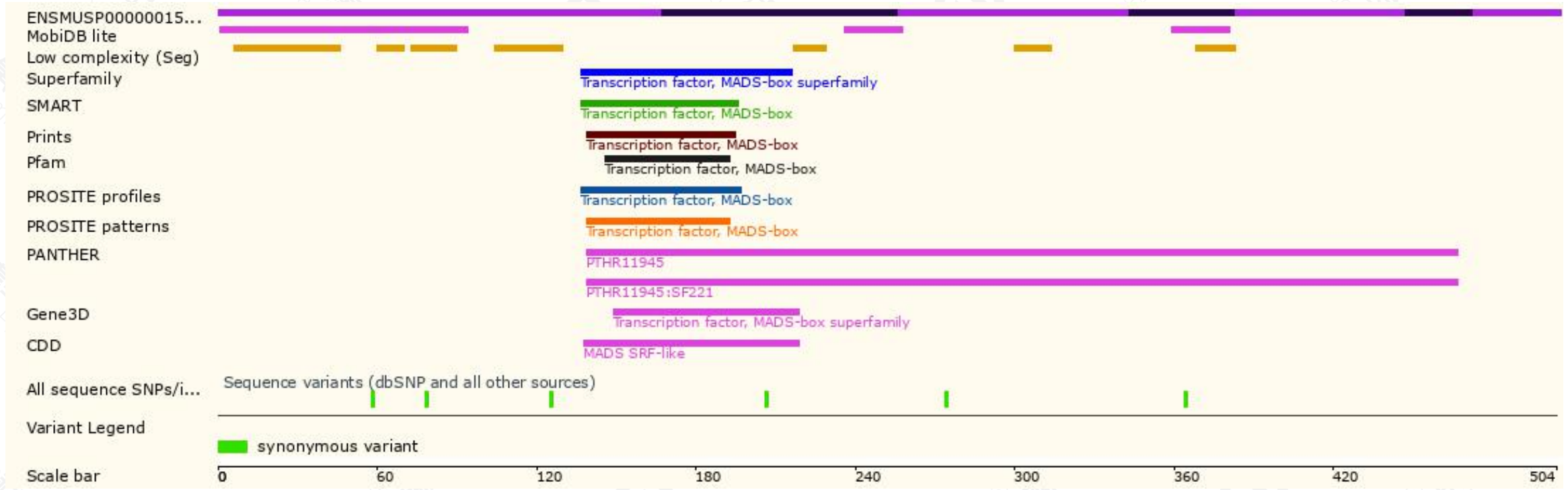
The strategy is based on the design of *Ptges2*-201 transcript, the transcription is shown below:



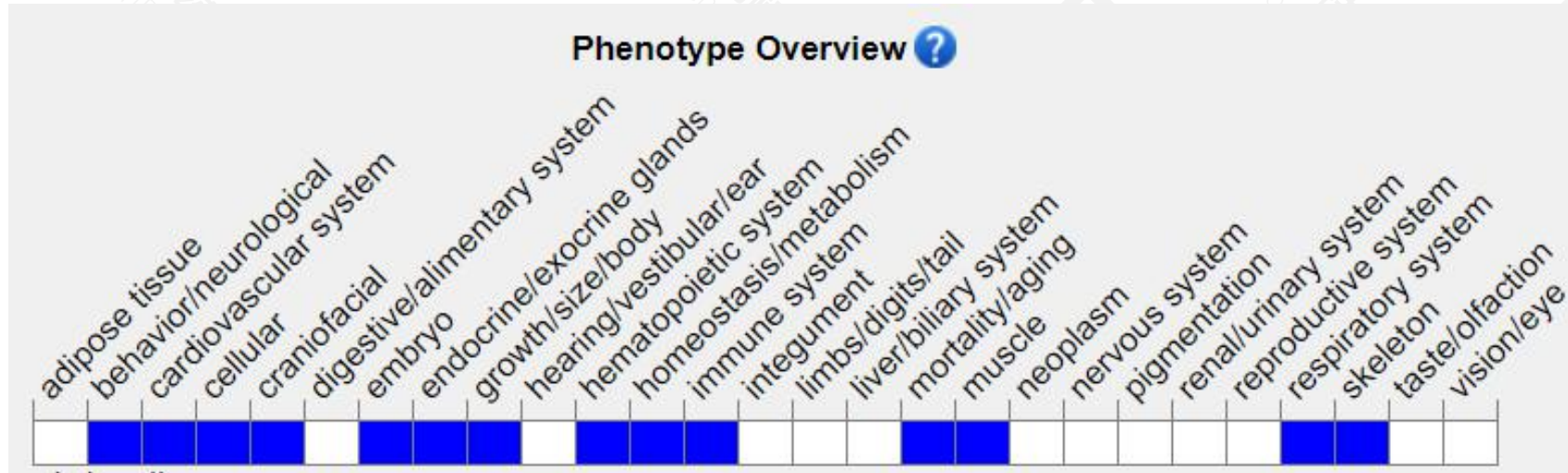
Genomic location distribution



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/>) .

Homozygous null mice exhibit embryonic lethality, abnormal gastrulation, no mesoderm or primitive streak formation and reduced embryo size.

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