

Six4 Cas9-CKO Strategy

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Project Overview

Project Name

Six4

Project type

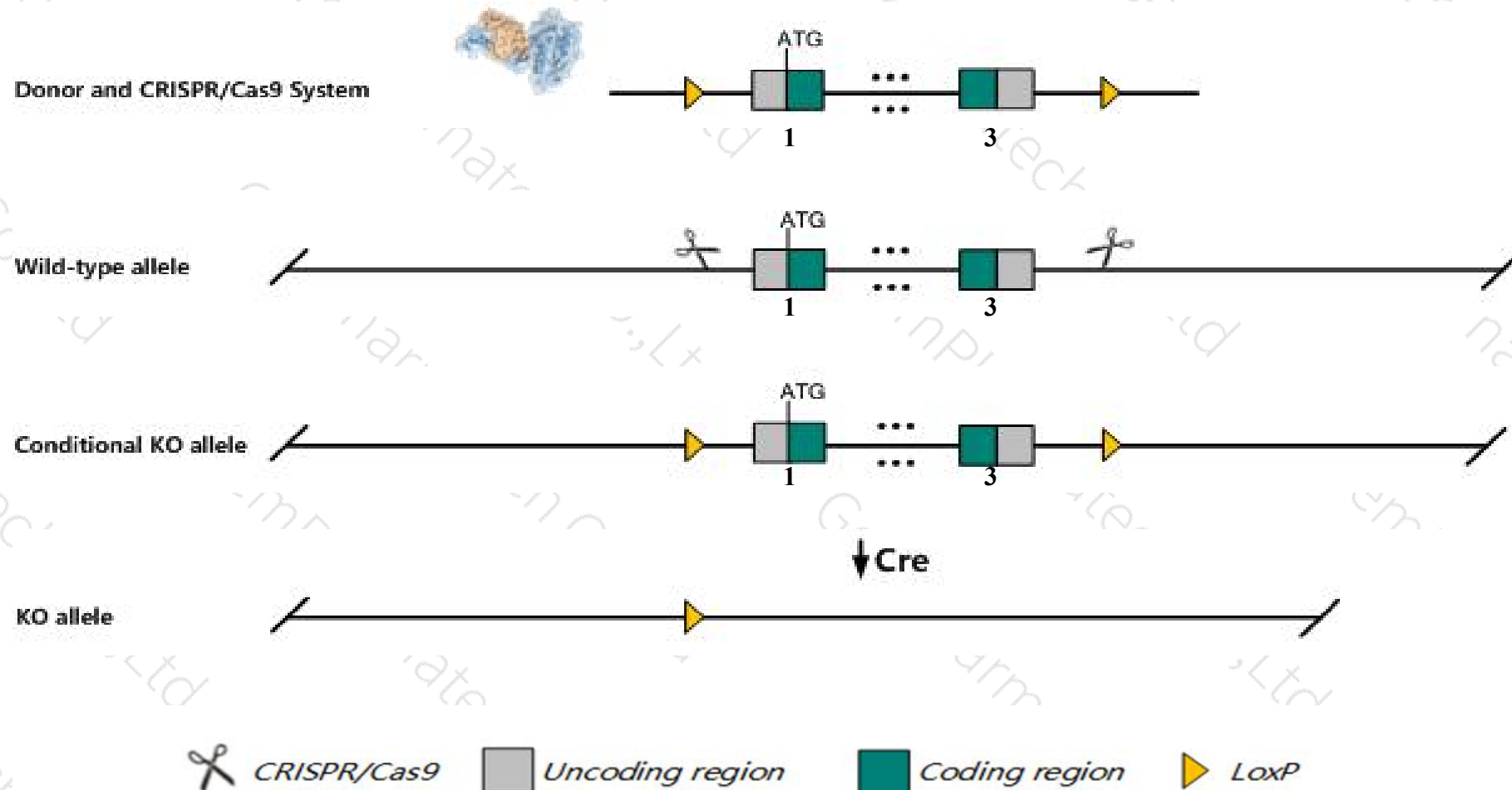
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Six4* gene. The schematic diagram is as follows:



- The *Six4* gene has 2 transcripts. According to the structure of *Six4* gene, exon1-exon3 of *Six4-201* (ENSMUST00000043208.7) transcript is recommended as the knockout region. The region contains all 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 *Six4* 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, Homozygotes for a targeted null mutation are viable, fertile, and exhibit no apparent abnormalities suggesting compensation by other Six family members.
- The *Six4* gene is located on the Chr12. 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)



Six4 sine oculis-related homeobox 4 [*Mus musculus* (house mouse)]

Gene ID: 20474, updated on 4-Dec-2019

Summary



Official Symbol	Six4 provided by MGI
Official Full Name	sine oculis-related homeobox 4 provided by MGI
Primary source	MGI:MGI:106034
See related	Ensembl:ENSMUSG00000034460
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	AREC3; TrexBF; AI047561
Expression	Broad expression in limb E14.5 (RPKM 2.1), CNS E11.5 (RPKM 1.4) and 15 other tissues See more
Orthologs	human all

Genomic context



Location: 12; 12 C3

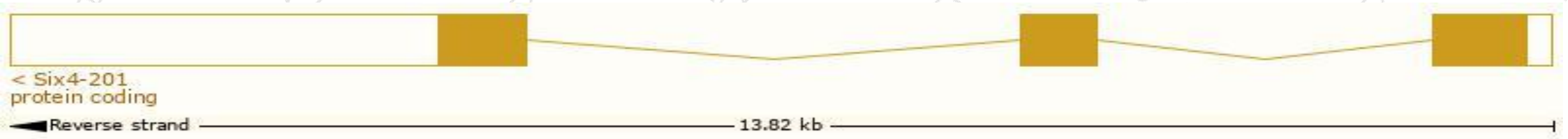
See Six4 in [Genome Data Viewer](#)

Transcript information (Ensembl)

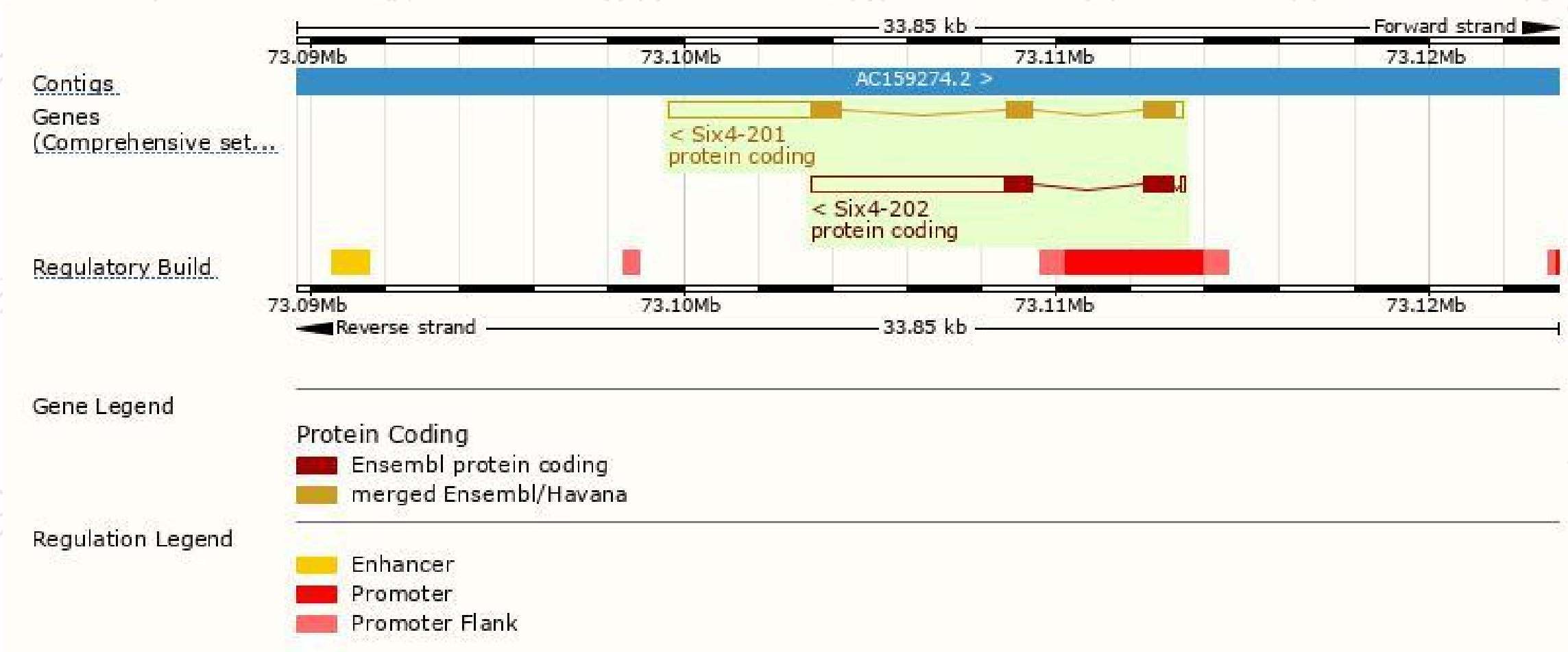
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Six4-201	ENSMUST00000043208.7	6402	775aa	Protein coding	CCDS25974	Q61321	TSL:1 GENCODE basic APPRIS P2
Six4-202	ENSMUST00000175693.2	6817	518aa	Protein coding	-	H3BL91	TSL:5 GENCODE basic APPRIS ALT2

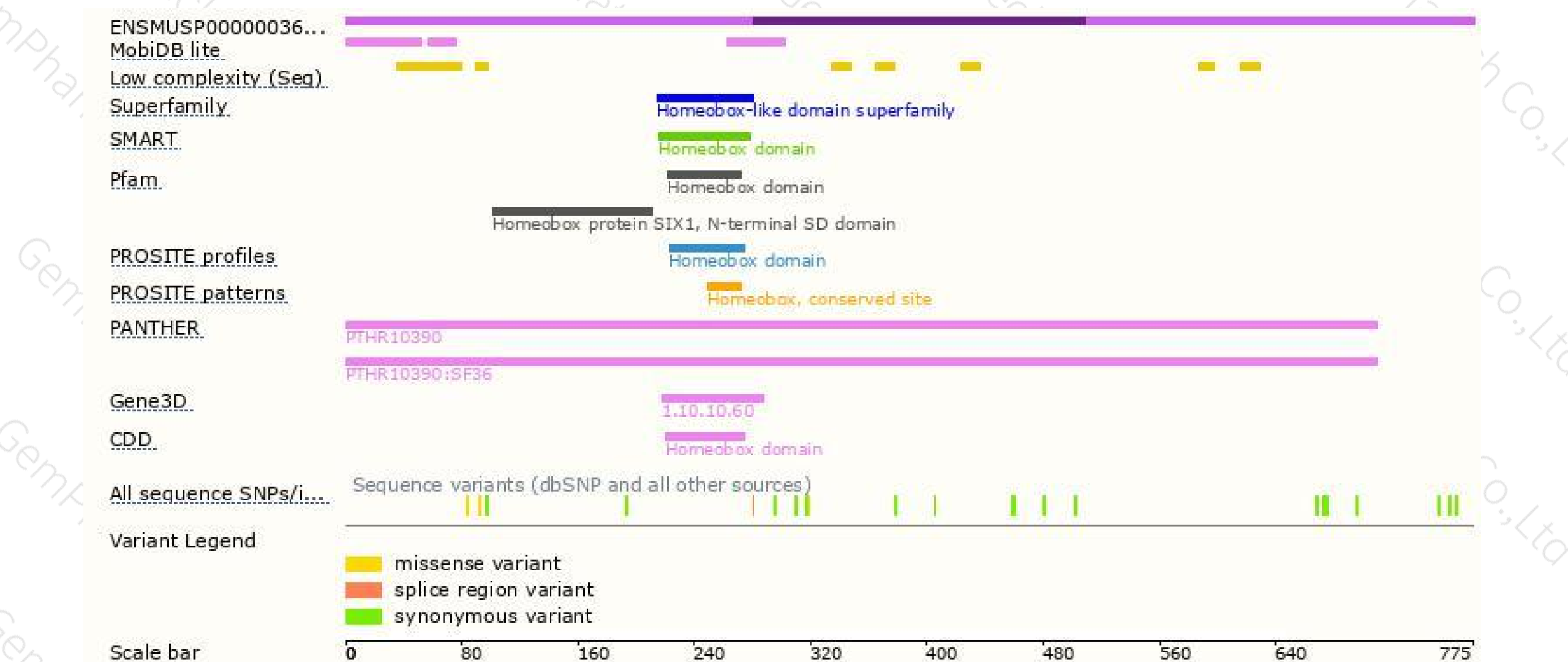
The strategy is based on the design of *Six4-201* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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