

# ***Klhl6* Cas9-KO Strategy**

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# Project Overview

**Project Name**

***Klhl6***

**Project type**

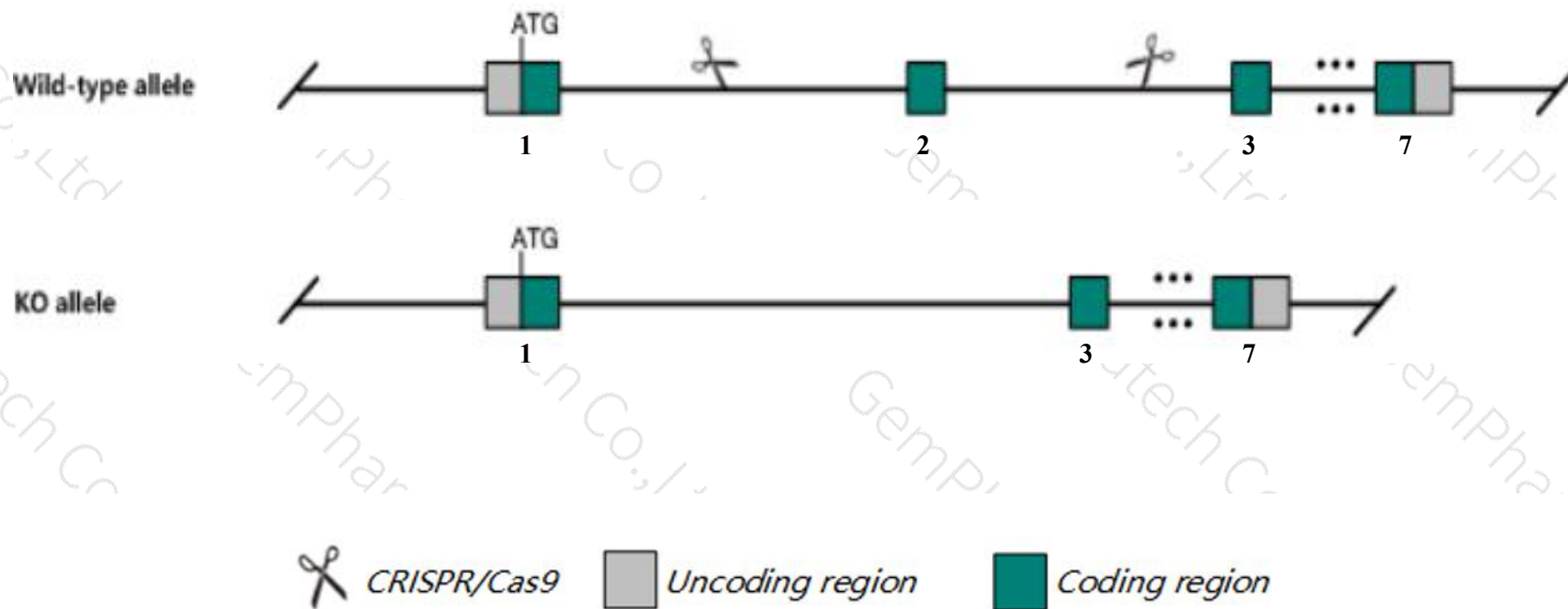
**Cas9-KO**

**Strain background**

**C57BL/6JGpt**

# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Klhl6* gene. The schematic diagram is as follows:



- The *Klhl6* gene has 4 transcripts. According to the structure of *Klhl6* gene, exon2 of *Klhl6-201*(ENSMUST00000058839.9) transcript is recommended as the knockout region. The region contains 166bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Klhl6* gene. The brief process is as follows: CRISPR/Cas9 system 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 knock-out allele exhibit spleen hypoplasia, defects in mature B-cell subsets with normal pro- and pre-B-cell development, severely impaired antigen-dependent germinal center formation, and reduced memory IgG response.
- The *Klhl6* gene is located on the Chr16. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.



# Gene information (NCBI)

## Klhl6 kelch-like 6 [Mus musculus (house mouse)]

Gene ID: 239743, updated on 13-Mar-2020

### Summary

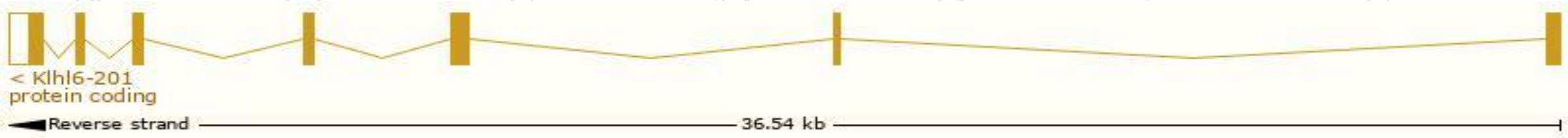
<b>Official Symbol</b>	Klhl6 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	kelch-like 6 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:2686922</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000043008</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	VALIDATED
<b>Organism</b>	<a href="#">Mus musculus</a>
<b>Lineage</b>	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
<b>Expression</b>	Broad expression in thymus adult (RPKM 18.2), spleen adult (RPKM 14.8) and 17 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

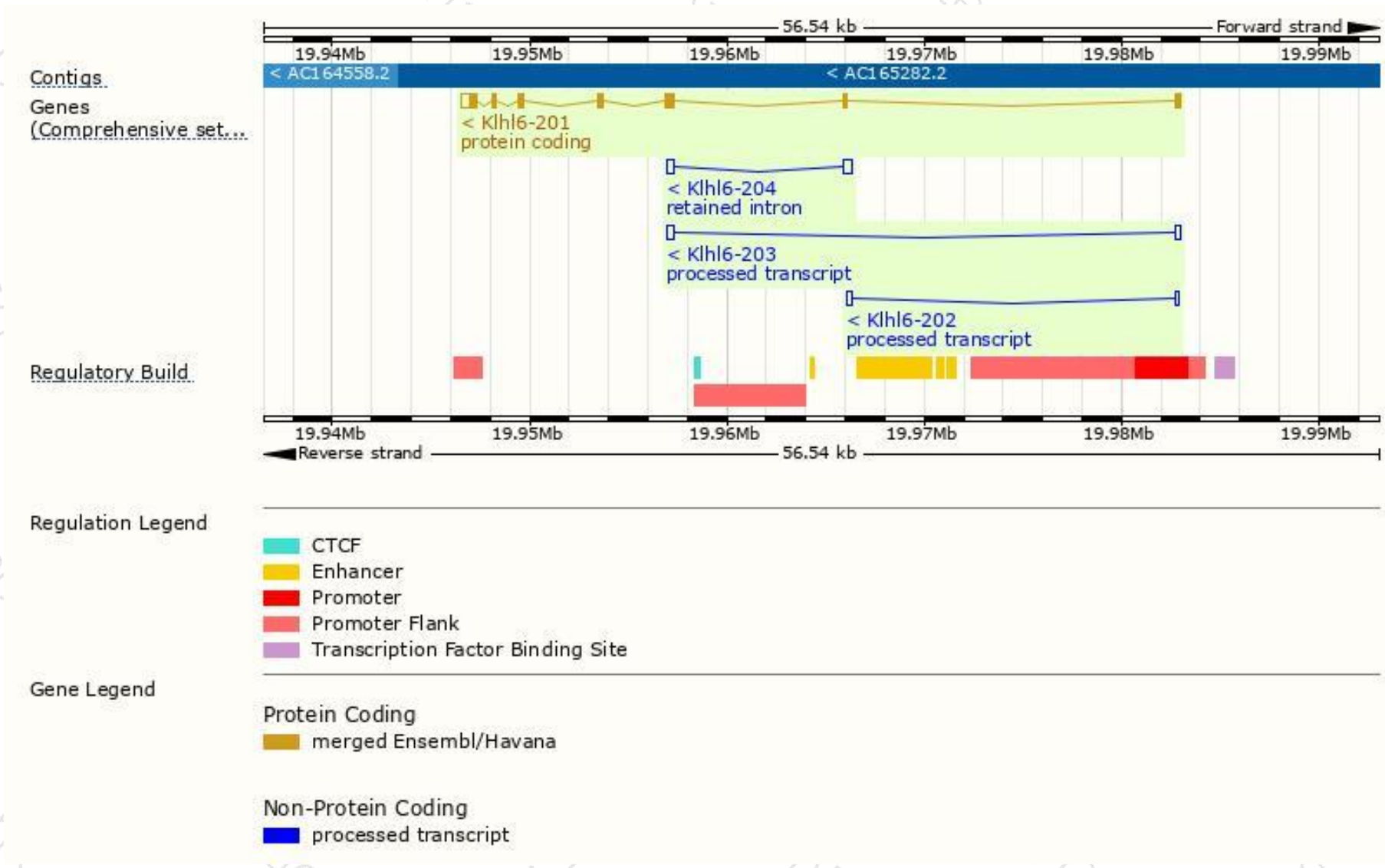
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Klhl6-201	<a href="#">ENSMUST00000058839.9</a>	2389	<a href="#">619aa</a>	Protein coding	<a href="#">CCDS28041</a>	<a href="#">Q6V595</a>	TSL:1 GENCODE basic APPRIS P1
Klhl6-203	<a href="#">ENSMUST00000166801.1</a>	675	No protein	Processed transcript	-	-	TSL:5
Klhl6-202	<a href="#">ENSMUST00000165530.1</a>	484	No protein	Processed transcript	-	-	TSL:2
Klhl6-204	<a href="#">ENSMUST00000171910.1</a>	790	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Klhl6-201* transcript,the transcription is shown below:

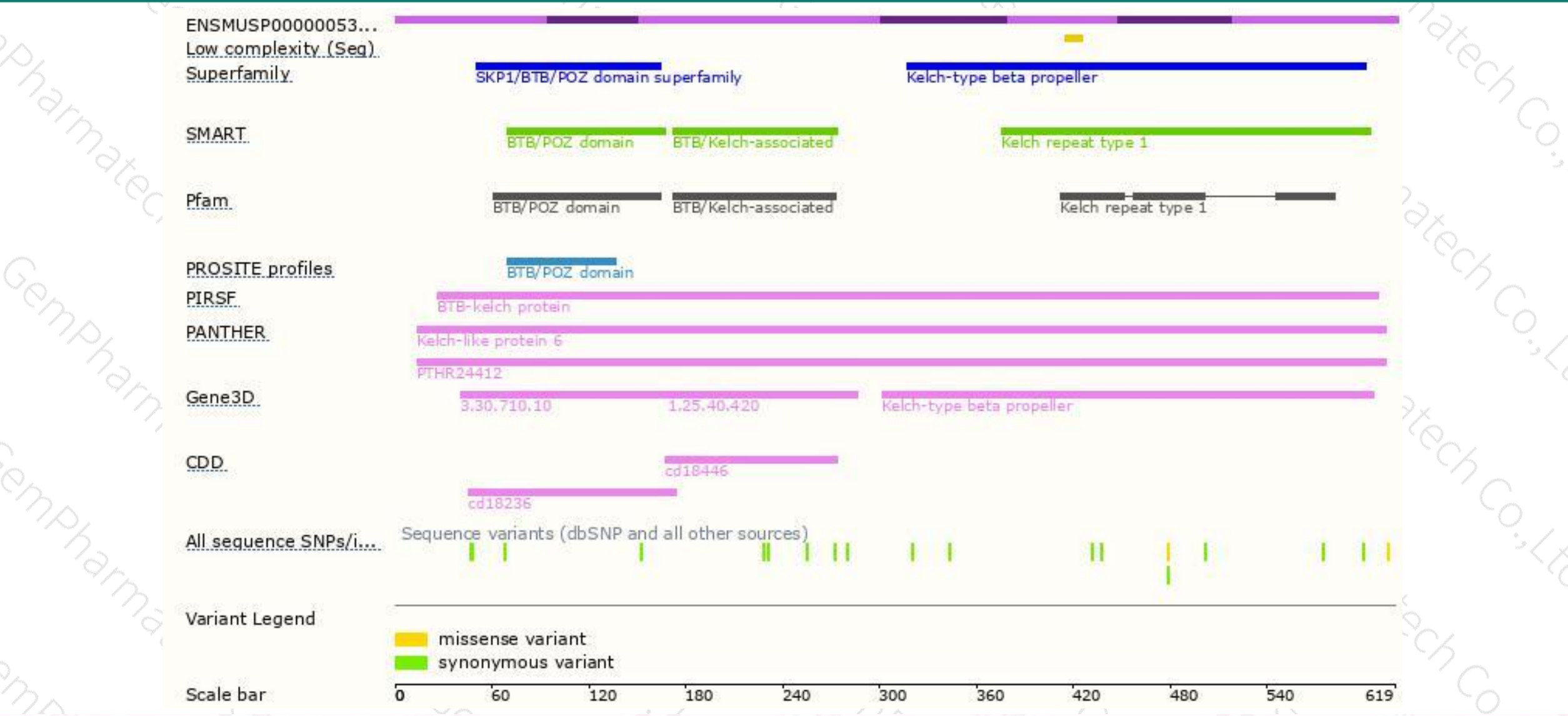


# Genomic location distribution

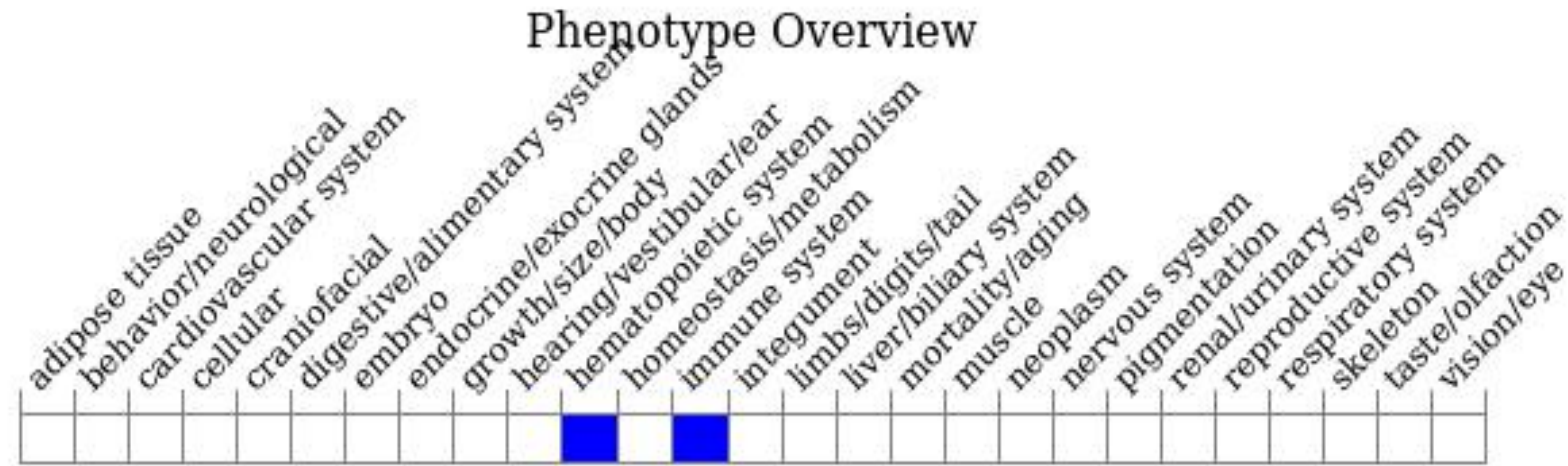




# Protein domain



# 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, mice homozygous for a knock-out allele exhibit spleen hypoplasia, defects in mature B-cell subsets with normal pro- and pre-B-cell development, severely impaired antigen-dependent germinal center formation, and reduced memory IgG response.

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

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