

# *Arhgef40* Cas9-CKO Strategy

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**Reviewer: Xiaojing Li**

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# Project Overview

**Project Name**

*Arhgef40*

**Project type**

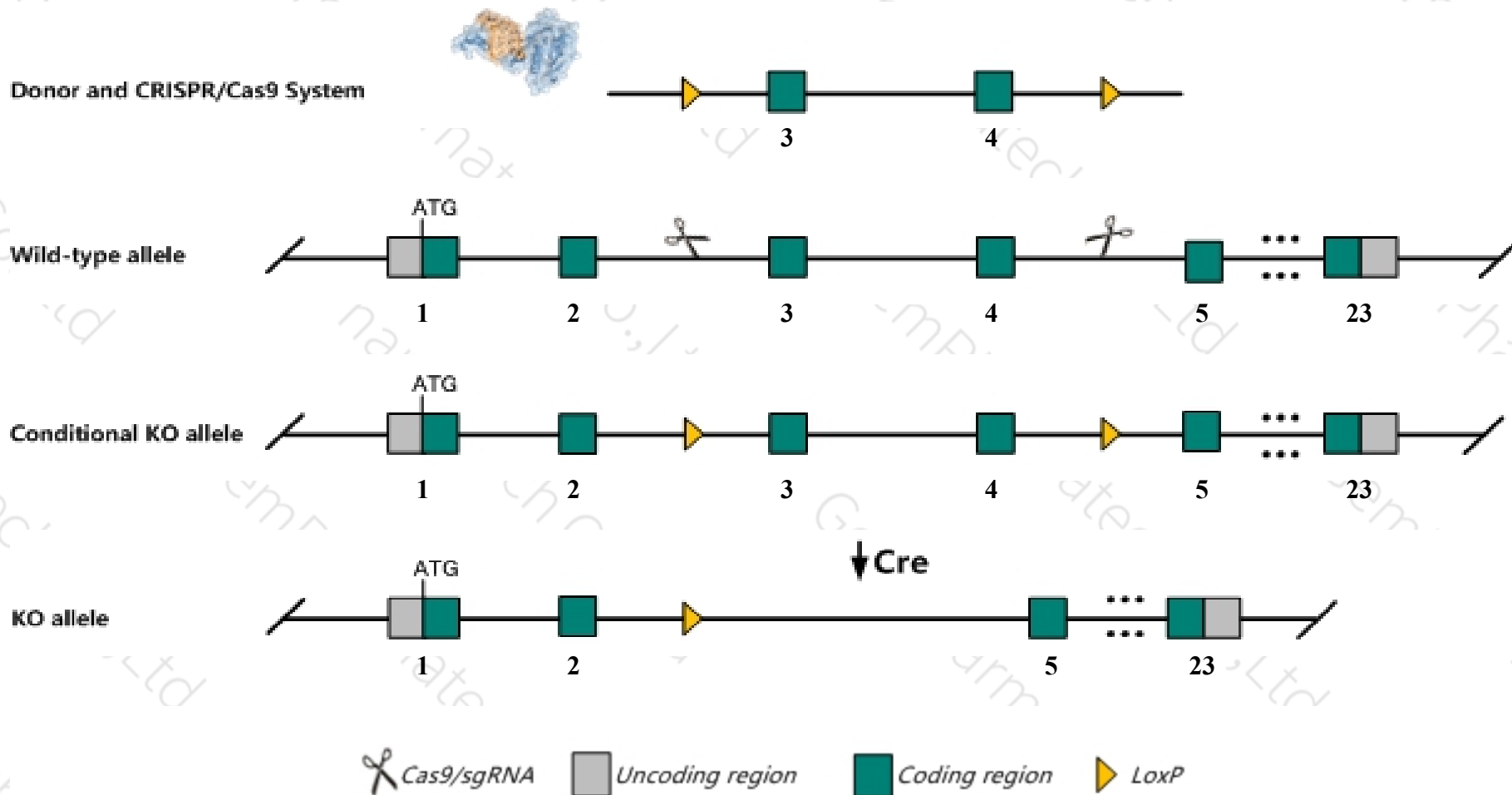
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Arhgef40* gene. The schematic diagram is as follows:



# Technical routes

- The *Arhgef40* gene has 26 transcripts. According to the structure of *Arhgef40* gene, exon3-exon4 of *Arhgef40-201*(ENSMUST00000093813.11) transcript is recommended as the knockout region. The region contains 1405bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Arhgef40* 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.
- The flox mice was 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.

- The *Arhgef40* gene is located on the Chr14. 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.
- Transcript 206,214,218 CDS 5' incomplete the influences is unknown.
- Transcript 205 CDS 3' incomplete the influences is unknown.
- 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)

## Arhgef40 Rho guanine nucleotide exchange factor (GEF) 40 [Mus musculus (house mouse)]

Gene ID: 268739, updated on 13-Mar-2020

### Summary



<b>Official Symbol</b>	Arhgef40 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	Rho guanine nucleotide exchange factor (GEF) 40 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:2685515</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000004562</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>Also known as</b>	E130112L23Rik, Gm669, Solo
<b>Expression</b>	Ubiquitous expression in limb E14.5 (RPKM 18.9), genital fat pad adult (RPKM 11.5) and 27 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

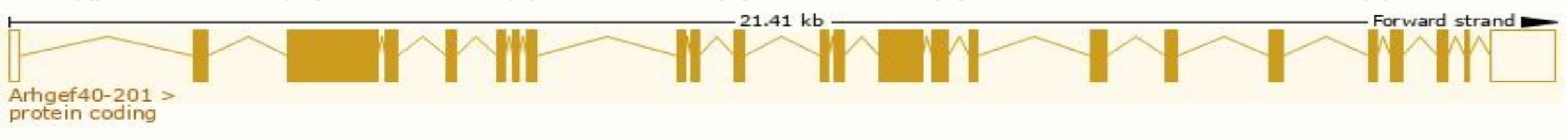


# Transcript information (Ensembl)

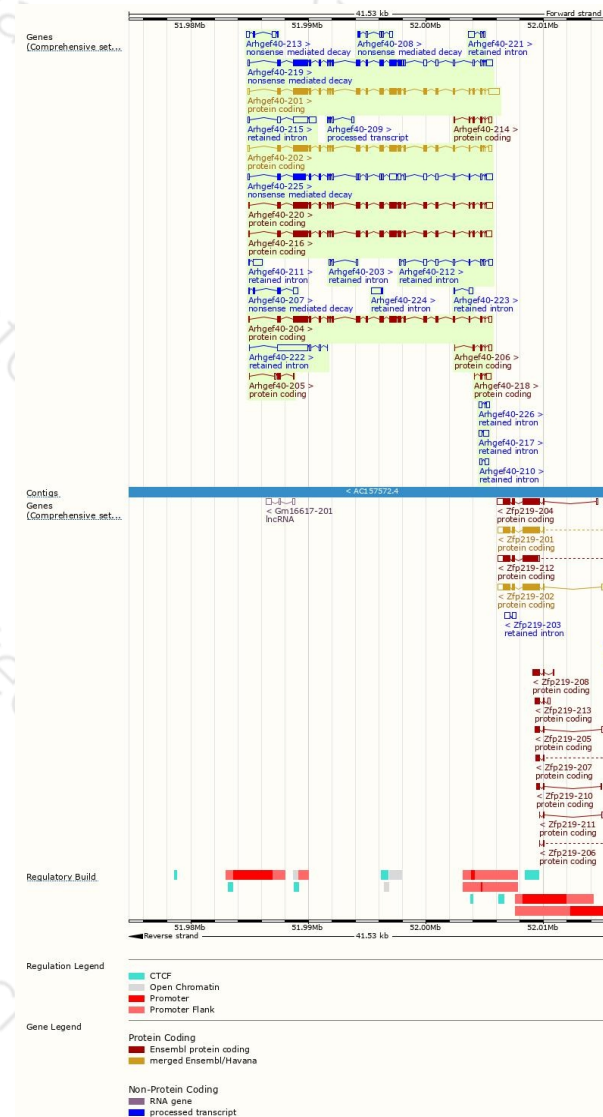
The gene has 26 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Arhgef40-201	<a href="#">ENSMUST00000093813.1</a>	5621	<a href="#">1517aa</a>	Protein coding	<a href="#">CCDS49488</a>	<a href="#">Q3UPH7</a>	TSL:1 GENCODE basic APPRIS P4
Arhgef40-220	<a href="#">ENSMUST00000182909.7</a>	5108	<a href="#">1517aa</a>	Protein coding	<a href="#">CCDS49488</a>	<a href="#">Q3UPH7</a>	TSL:5 GENCODE basic APPRIS P4
Arhgef40-202	<a href="#">ENSMUST00000100639.10</a>	4843	<a href="#">1476aa</a>	Protein coding	<a href="#">CCDS49489</a>	<a href="#">Q3UPH7</a>	TSL:1 GENCODE basic APPRIS ALT2
Arhgef40-204	<a href="#">ENSMUST00000182061.7</a>	4756	<a href="#">1469aa</a>	Protein coding	<a href="#">CCDS59616</a>	<a href="#">Q3UPH7</a>	TSL:1 GENCODE basic APPRIS ALT2
Arhgef40-216	<a href="#">ENSMUST00000182760.7</a>	5130	<a href="#">1526aa</a>	Protein coding	-	<a href="#">S4R189</a>	TSL:5 GENCODE basic APPRIS ALT2
Arhgef40-214	<a href="#">ENSMUST00000182667.7</a>	827	<a href="#">179aa</a>	Protein coding	-	<a href="#">S4R1Y4</a>	CDS 5' incomplete TSL:3
Arhgef40-206	<a href="#">ENSMUST00000182217.7</a>	638	<a href="#">115aa</a>	Protein coding	-	<a href="#">S4R2A6</a>	CDS 5' incomplete TSL:5
Arhgef40-218	<a href="#">ENSMUST00000182828.1</a>	608	<a href="#">73aa</a>	Protein coding	-	<a href="#">S4R1R5</a>	CDS 5' incomplete TSL:3
Arhgef40-205	<a href="#">ENSMUST00000182193.1</a>	393	<a href="#">89aa</a>	Protein coding	-	<a href="#">S4R2B3</a>	CDS 3' incomplete TSL:5
Arhgef40-219	<a href="#">ENSMUST00000182905.7</a>	5320	<a href="#">1191aa</a>	Nonsense mediated decay	-	<a href="#">S4R2U9</a>	TSL:1
Arhgef40-225	<a href="#">ENSMUST00000183208.7</a>	4826	<a href="#">500aa</a>	Nonsense mediated decay	-	<a href="#">S4R1S6</a>	TSL:5
Arhgef40-208	<a href="#">ENSMUST00000182412.1</a>	822	<a href="#">45aa</a>	Nonsense mediated decay	-	<a href="#">S4R1X0</a>	CDS 5' incomplete TSL:3
Arhgef40-207	<a href="#">ENSMUST00000182338.1</a>	812	<a href="#">85aa</a>	Nonsense mediated decay	-	<a href="#">S4R237</a>	TSL:3
Arhgef40-213	<a href="#">ENSMUST00000182649.7</a>	751	<a href="#">44aa</a>	Nonsense mediated decay	-	<a href="#">S4R2G8</a>	TSL:5
Arhgef40-209	<a href="#">ENSMUST00000182480.7</a>	461	No protein	Processed transcript	-	-	TSL:3
Arhgef40-222	<a href="#">ENSMUST00000182961.1</a>	2903	No protein	Retained intron	-	-	TSL:1
Arhgef40-215	<a href="#">ENSMUST00000182740.1</a>	2191	No protein	Retained intron	-	-	TSL:1
Arhgef40-212	<a href="#">ENSMUST00000182644.7</a>	1429	No protein	Retained intron	-	-	TSL:1
Arhgef40-224	<a href="#">ENSMUST00000183167.1</a>	924	No protein	Retained intron	-	-	TSL:3
Arhgef40-211	<a href="#">ENSMUST00000182609.1</a>	848	No protein	Retained intron	-	-	TSL:3
Arhgef40-221	<a href="#">ENSMUST00000182931.1</a>	820	No protein	Retained intron	-	-	TSL:5
Arhgef40-217	<a href="#">ENSMUST00000182777.1</a>	676	No protein	Retained intron	-	-	TSL:3
Arhgef40-226	<a href="#">ENSMUST00000183213.1</a>	515	No protein	Retained intron	-	-	TSL:2
Arhgef40-210	<a href="#">ENSMUST00000182504.1</a>	478	No protein	Retained intron	-	-	TSL:3
Arhgef40-223	<a href="#">ENSMUST00000183022.1</a>	387	No protein	Retained intron	-	-	TSL:3
Arhgef40-203	<a href="#">ENSMUST00000182019.1</a>	336	No protein	Retained intron	-	-	TSL:3

The strategy is based on the design of *Arhgef40-201* transcript,the transcription is shown below:

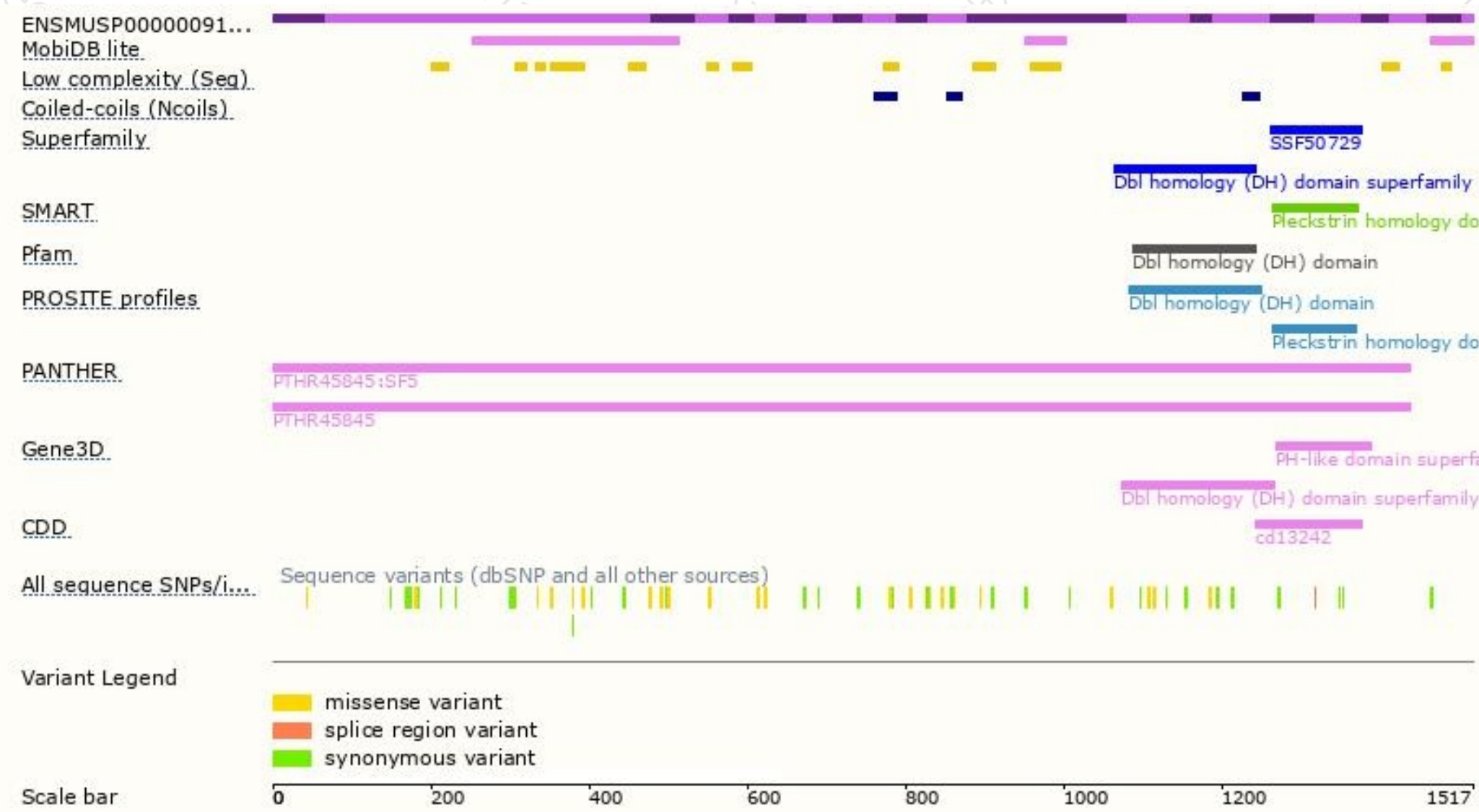


# Genomic location distribution





# Protein domain



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

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