

Rasal3 Cas9-CKO Strategy

Designer:

Project Overview

Project Name

Rasal3

Project type

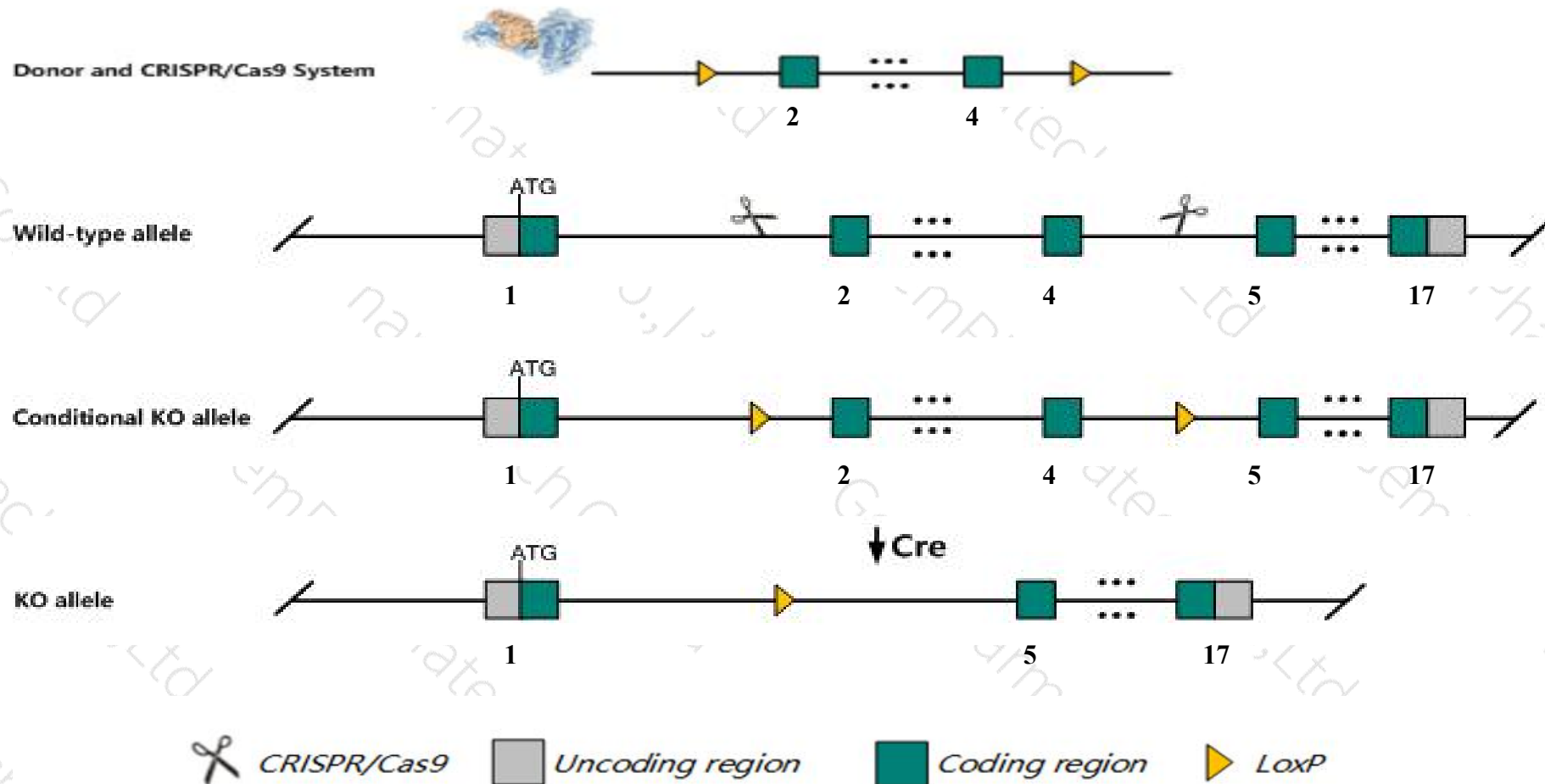
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rasal3* gene. The schematic diagram is as follows:



- The *Rasal3* gene has 10 transcripts. According to the structure of *Rasal3* gene, exon2-exon4 of *Rasal3-201* (ENSMUST00000063824.13) transcript is recommended as the knockout region. The region contains 281bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rasal3* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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, Mice homozygous for a knock-out allele exhibit reduced NT T cells in the liver, increased granulocytes in the bone marrow and decreased susceptibility to alpha-GalCer-induced liver injury.
- The *Rasal3* gene is located on the Chr17. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information (NCBI)

Rasal3 RAS protein activator like 3 [Mus musculus (house mouse)]

Gene ID: 320484, updated on 31-Jan-2019

Summary



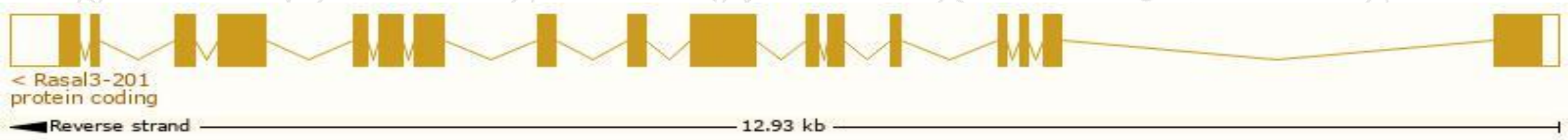
Official Symbol	Rasal3 provided by MGI
Official Full Name	RAS protein activator like 3 provided by MGI
Primary source	MGI:MGI:2444128
See related	Ensembl:ENSMUSG00000052142
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	A430107D22Rik
Expression	Biased expression in thymus adult (RPKM 113.1), spleen adult (RPKM 50.6) and 1 other tissue See more
Orthologs	human all

Transcript information (Ensembl)

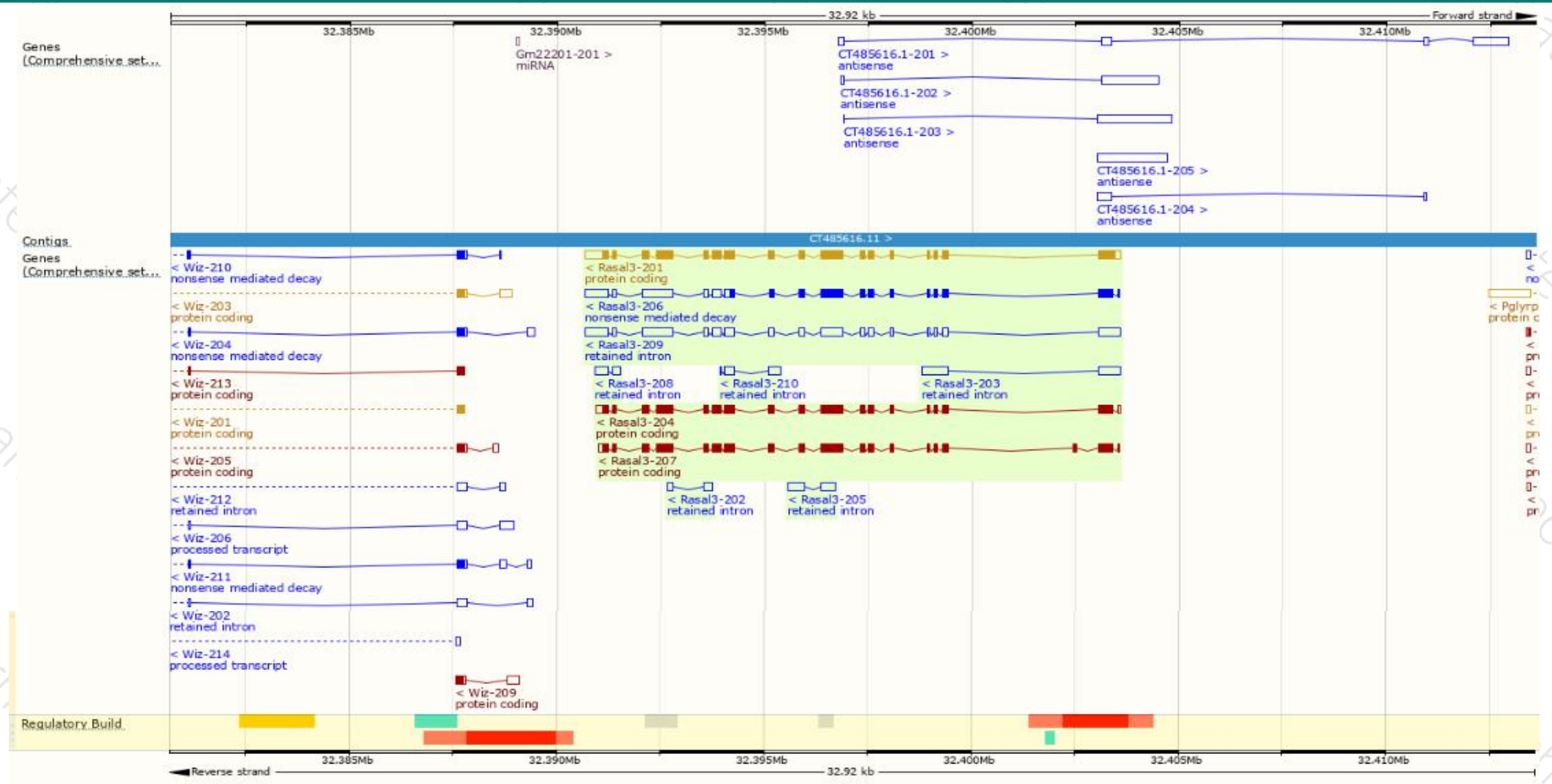
The gene has 10 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rasal3-201	ENSMUST00000063824.13	3681	1041aa	Protein coding	CCDS37556	Q8C2K5	TSL:1 GENCODE basic APPRIS P3
Rasal3-207	ENSMUST00000137458.1	3272	1043aa	Protein coding	CCDS84292	Q8C2K5	TSL:1 GENCODE basic APPRIS ALT2
Rasal3-204	ENSMUST00000135618.7	3293	1019aa	Protein coding	-	D3Z6Z7	TSL:5 GENCODE basic APPRIS ALT2
Rasal3-206	ENSMUST00000136375.7	3691	601aa	Nonsense mediated decay	-	A0A0R4J1U7	TSL:2
Rasal3-209	ENSMUST00000142203.7	3849	No protein	Retained intron	-	-	TSL:2
Rasal3-203	ENSMUST00000135560.1	1156	No protein	Retained intron	-	-	TSL:1
Rasal3-205	ENSMUST00000135968.1	762	No protein	Retained intron	-	-	TSL:2
Rasal3-210	ENSMUST00000143808.1	554	No protein	Retained intron	-	-	TSL:2
Rasal3-208	ENSMUST00000141714.1	492	No protein	Retained intron	-	-	TSL:2
Rasal3-202	ENSMUST00000134723.1	354	No protein	Retained intron	-	-	TSL:1

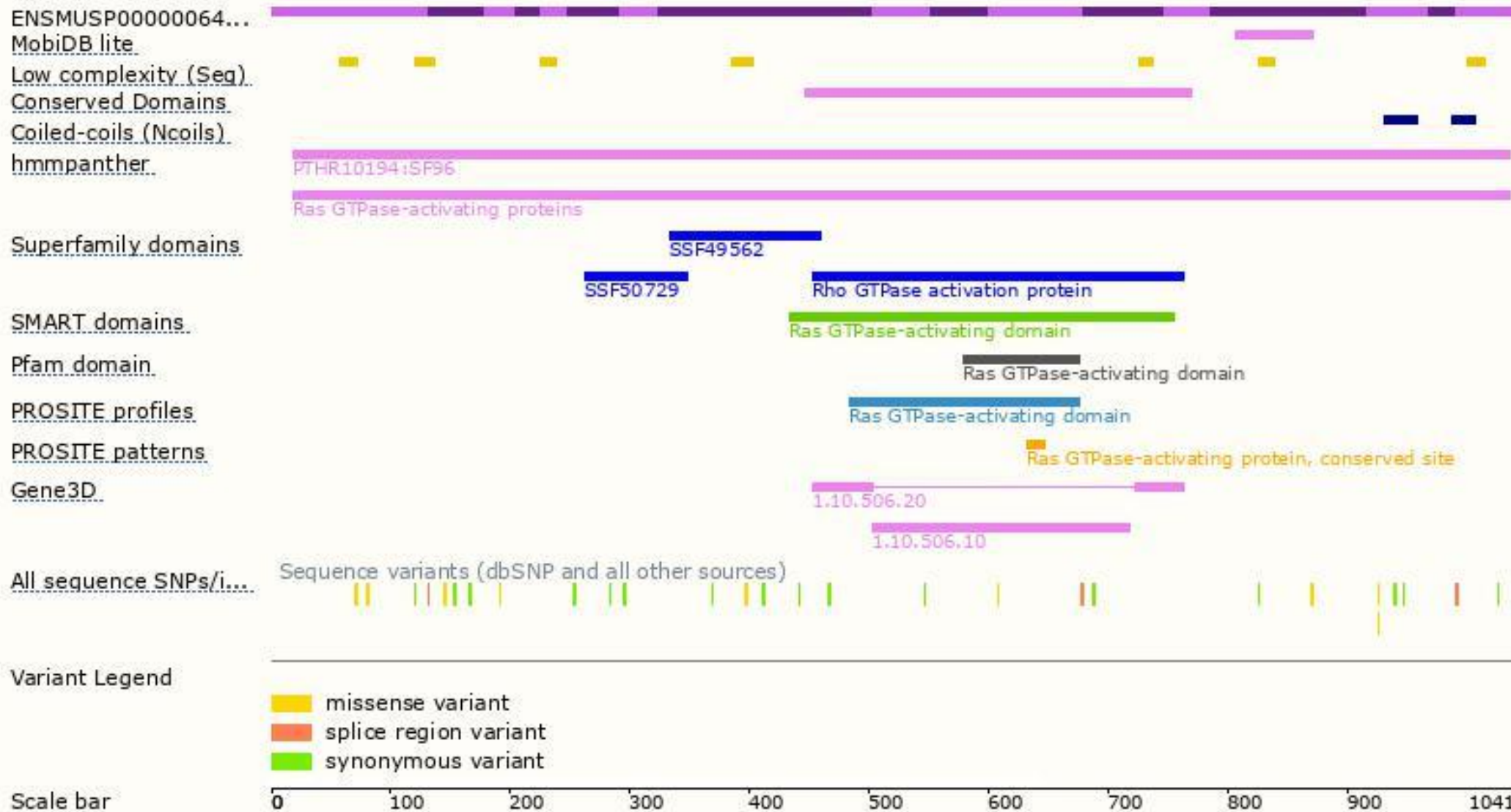
The strategy is based on the design of *Rasal3-201* transcript,The transcription is shown below



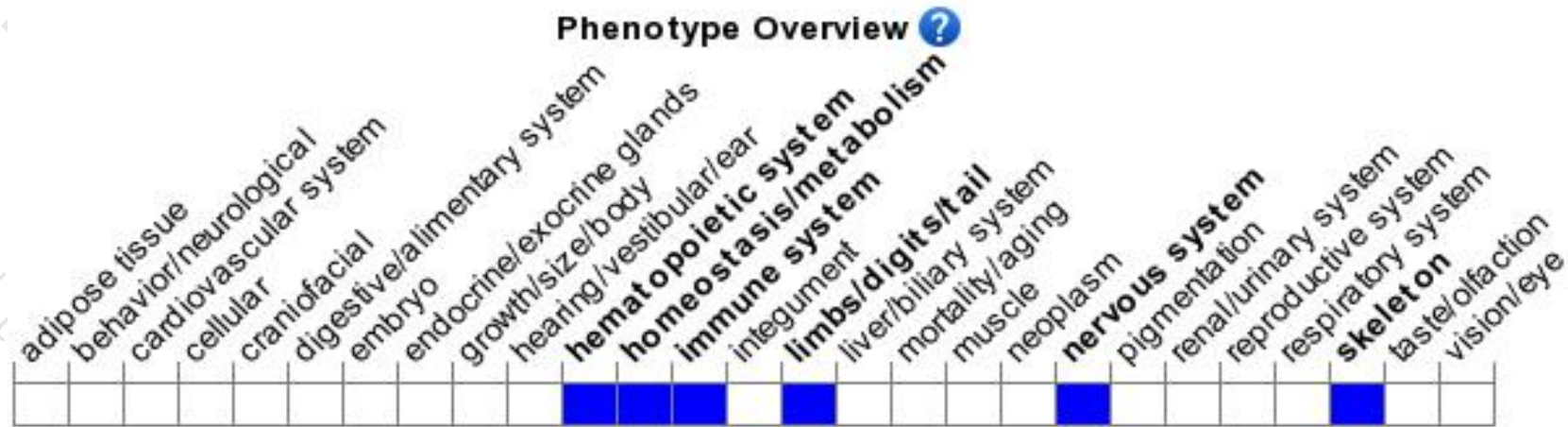
Genomic location distribution



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 reduced NT T cells in the liver, increased granulocytes in the bone marrow and decreased susceptibility to alpha-GalCer-induced liver injury.

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

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