Il2ra Cas9-KO Strategy

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Reviewer: Xiaojing Li

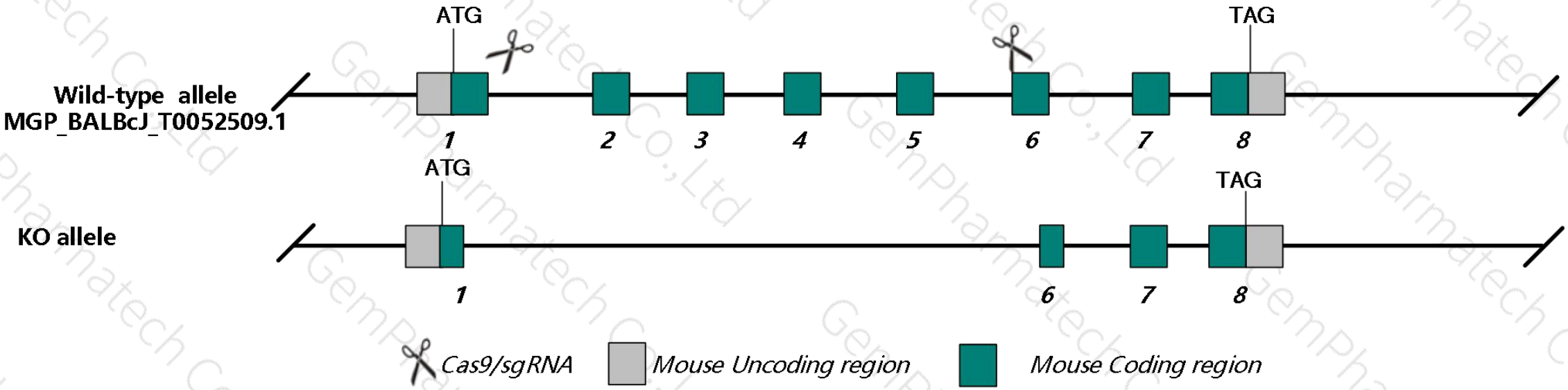
Design Date: 2020-7-24

Project Overview

Project Name	***Il2ra***
Project type	**Cas9-KO**
Strain background	**BALB/cJGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Il2ra* gene. The schematic diagram is as follows:



Technical routes

➢ The *Il2ra* gene has 3 transcripts. According to the structure of *Il2ra* gene, exon1-exon6 of MGP_BALBcJ_T0052509.1transcript is recommended as the knockout region.Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Il2ra* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of BALB/cJGpt 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 BALB/cJGpt mice.

Notice

➢ According to the existing MGI data,homozygotes for a targeted null mutation exhibit massive proliferation of polyclonal T and B cells as adults and develop autoimmune disorders including inflammatory bowel disease and hemolytic anemia with age.

➢ The *Il2ra* gene is located on the Chr2. 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）

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Il2ra interleukin 2 receptor, alpha chain [Mus musculus (house mouse)]

Gene ID: 16184, updated on 13-Mar-2020

Summary

Official Symbol	Il2ra provided by MGI
Official Full Name	interleukin 2 receptor, alpha chain provided by MGI
Primary source	MGI:MGI:96549
See related	Ensembl:ENSMUSG00000026770
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	CD25, Il2r, Ly-43
Expression	Biased expression in thymus adult (RPKM 5.6), mammary gland adult (RPKM 1.1) and 2 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 3 transcripts,all transcripts are shown below:

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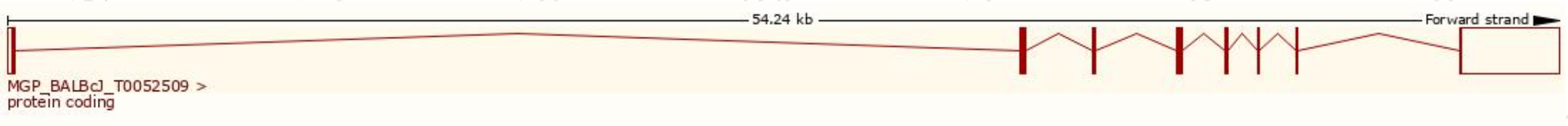
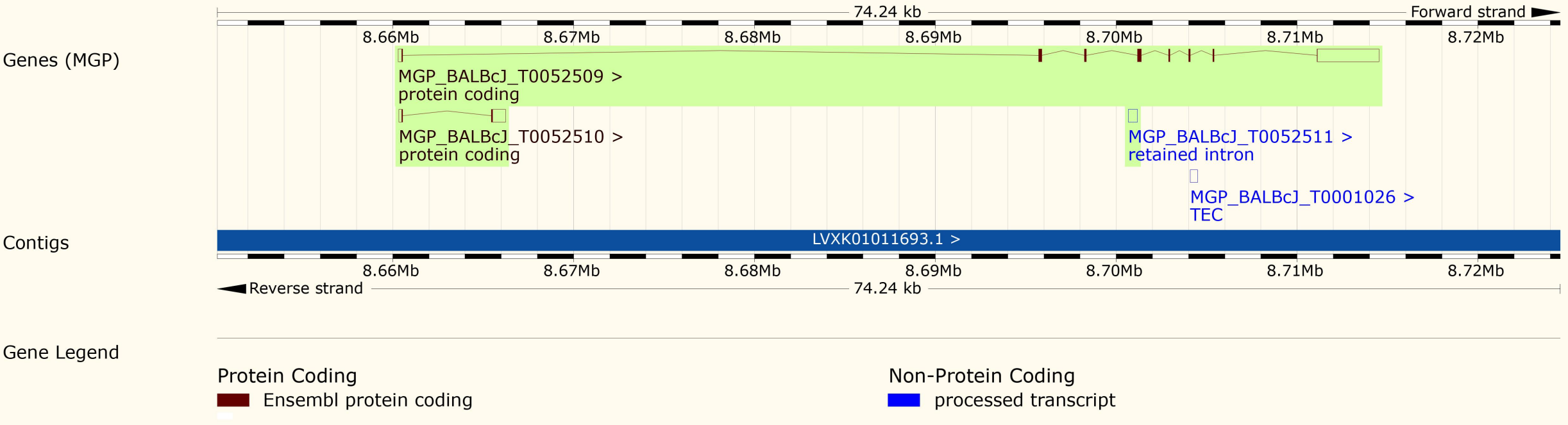
Name ▲	Transcript ID	bp	Protein	Translation ID	Biotype	CCDS	UniProt	Flags
-	MGP_BALBcJ_T0052509.1	4412	268aa	MGP_BALBcJ_P0052509	Protein coding	CCDS15685	P01590 Q3THW2 Q544I2	-
-	MGP_BALBcJ_T0052510.1	1013	44aa	MGP_BALBcJ_P0052510	Protein coding	-	-	-
-	MGP_BALBcJ_T0052511.1	502	No protein	-	Retained intron	-	-	-

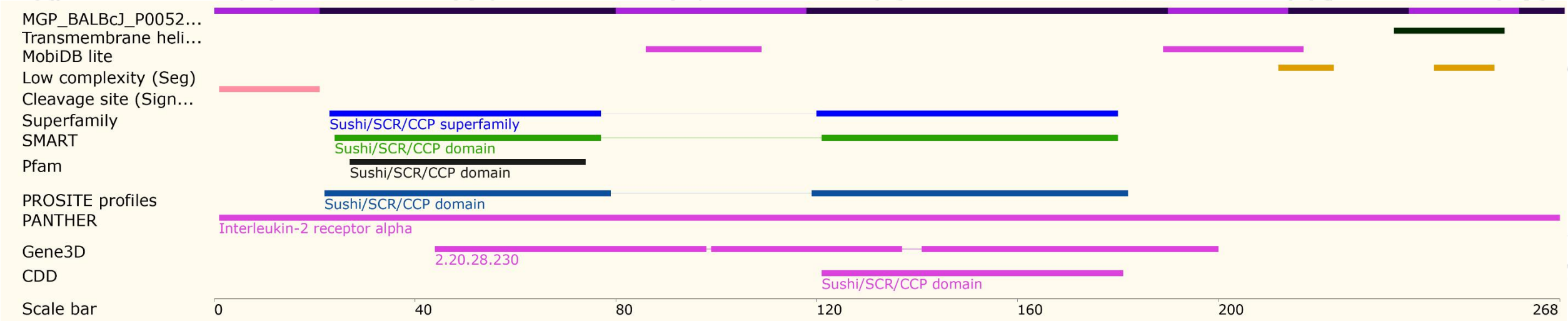
The strategy is based on the design of MGP_BALBcJ_T0052509.1 transcript,the transcription is shown below:



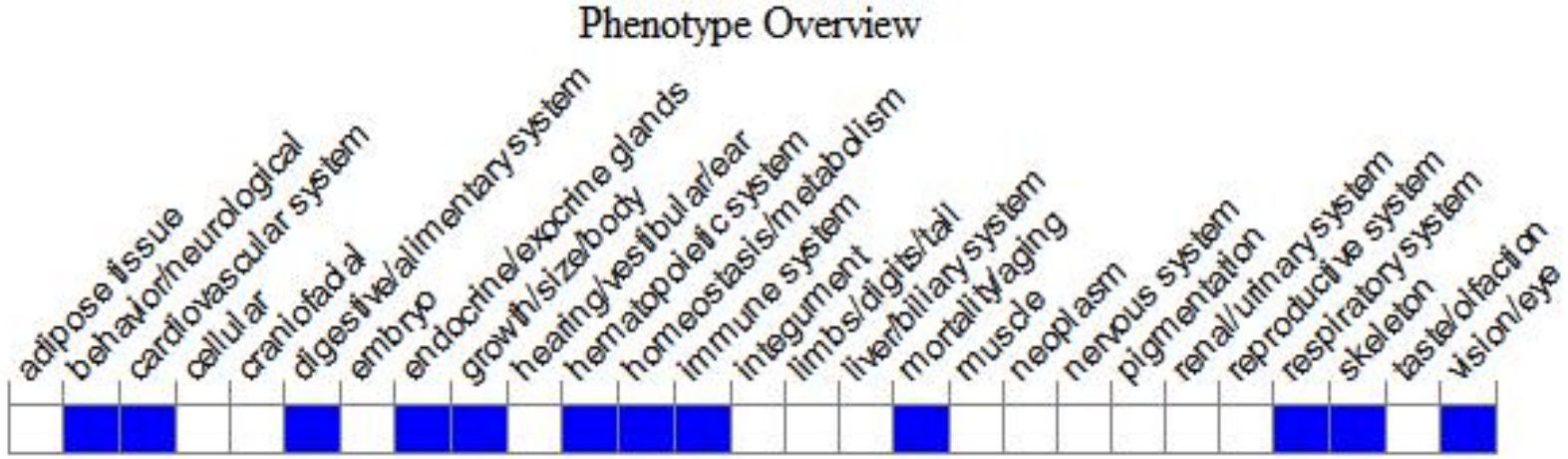
Genomic location distribution



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,homozygotes for a targeted null mutation exhibit massive proliferation of polyclonal T and B cells as adults and develop autoimmune disorders including inflammatory bowel disease and hemolytic anemia with age.

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

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