Tlx2 Cas9-KO Strategy

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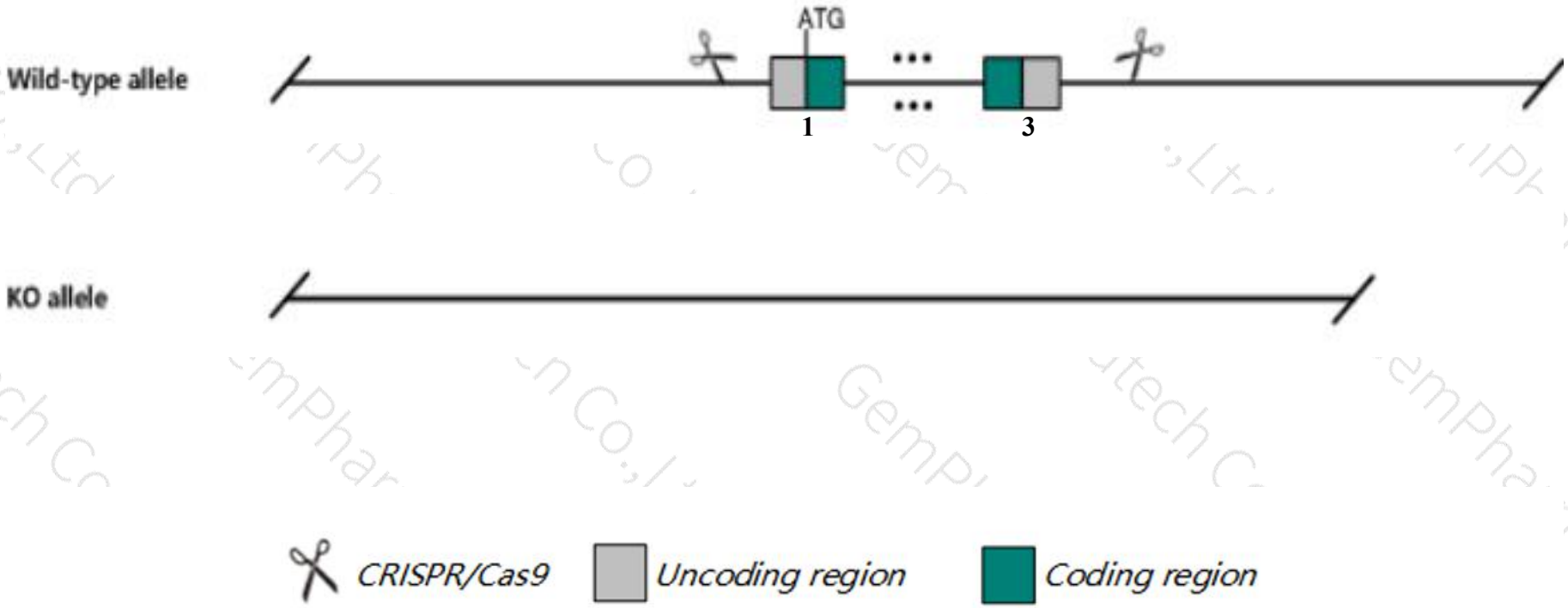
Design Date: 2020-7-20

Project Overview

Project Name	***Tlx2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tlx2* gene. The schematic diagram is as follows:



Technical routes

➢ The *Tlx2* gene has 2 transcripts. According to the structure of *Tlx2* gene, exon1-exon3 of *Tlx2-201*(ENSMUST00000089641.5) 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 *Tlx2* 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.

Notice

➢ According to the existing MGI data,homozygotes for two targeted null mutations exhibit megacolon with hyperinnervated enteric ganglia which undergo neuronal degeneration. Homozygotes for a third null mutation die in utero with defects in formation of the primitive streak and mesoderm.

➢ The *Tlx2* gene is located on the Chr6. 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）

集萃药康 GemPharmatech

Tlx2 T cell leukemia, homeobox 2 [Mus musculus (house mouse)]

Gene ID: 21909, updated on 13-Mar-2020

Summary

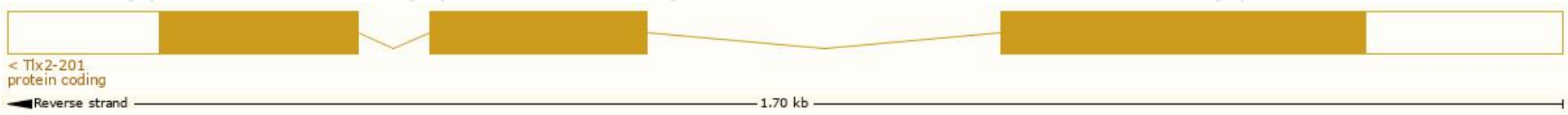
Official Symbol	Tlx2 provided by MGI
Official Full Name	T cell leukemia, homeobox 2 provided by MGI
Primary source	MGI:MGI:1350935
See related	Ensembl:ENSMUSG00000068327
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Enx, Hox11L.1, Hox11l1, NCX, Ncx1, Tlx1l1, Tlx1l2
Expression	Biased expression in adrenal adult (RPKM 24.6), duodenum adult (RPKM 3.7) and 4 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

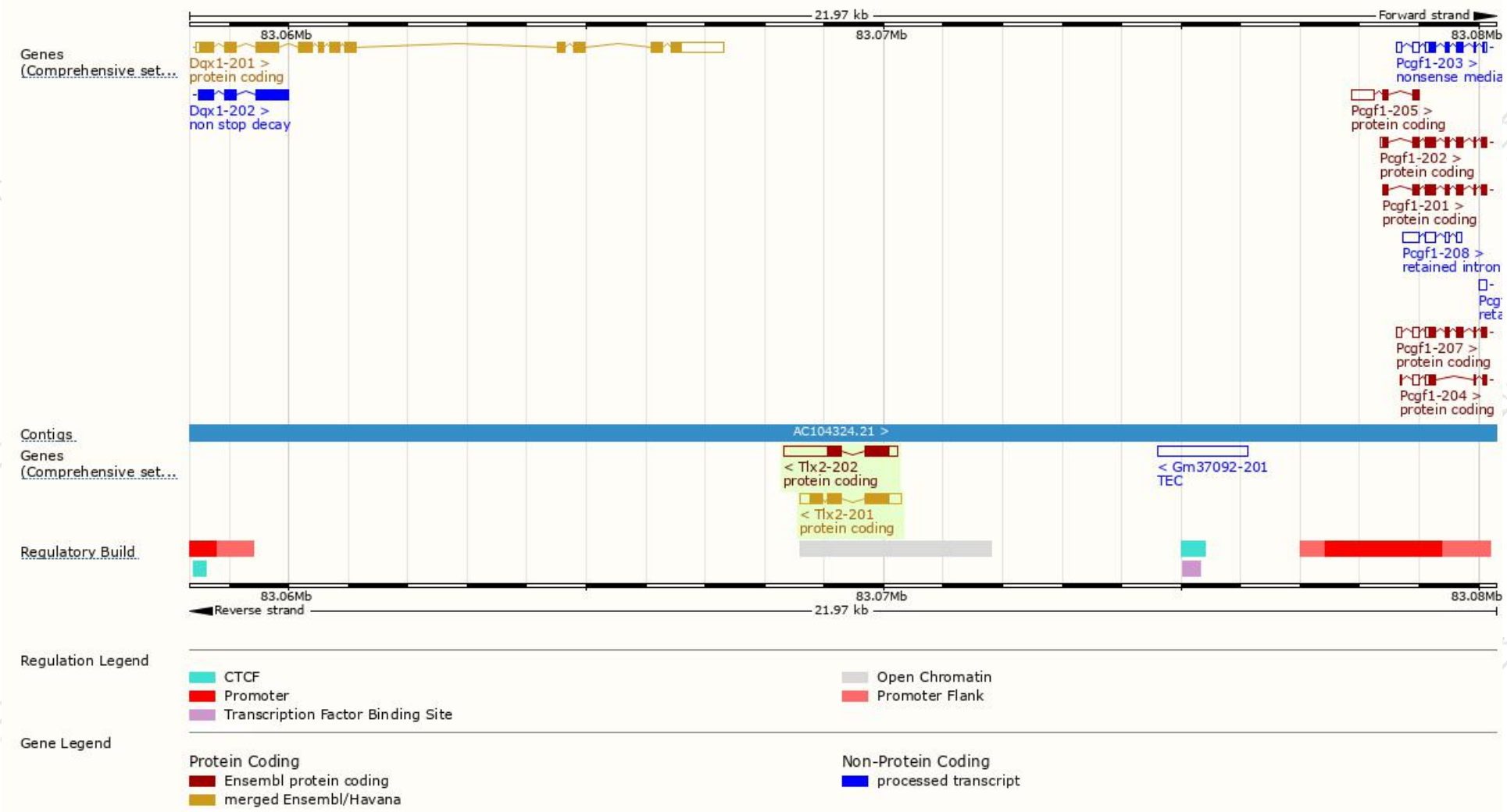
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein ▲	Biotype	CCDS	UniProt	Flags
Tlx2-202	ENSMUST00000174674.2	1514	213aa	Protein coding	-	G3UWU0	TSL:1 GENCODE basic
Tlx2-201	ENSMUST00000089641.5	1236	284aa	Protein coding	CCDS20269	Q61663	TSL:1 GENCODE basic APPRIS P1

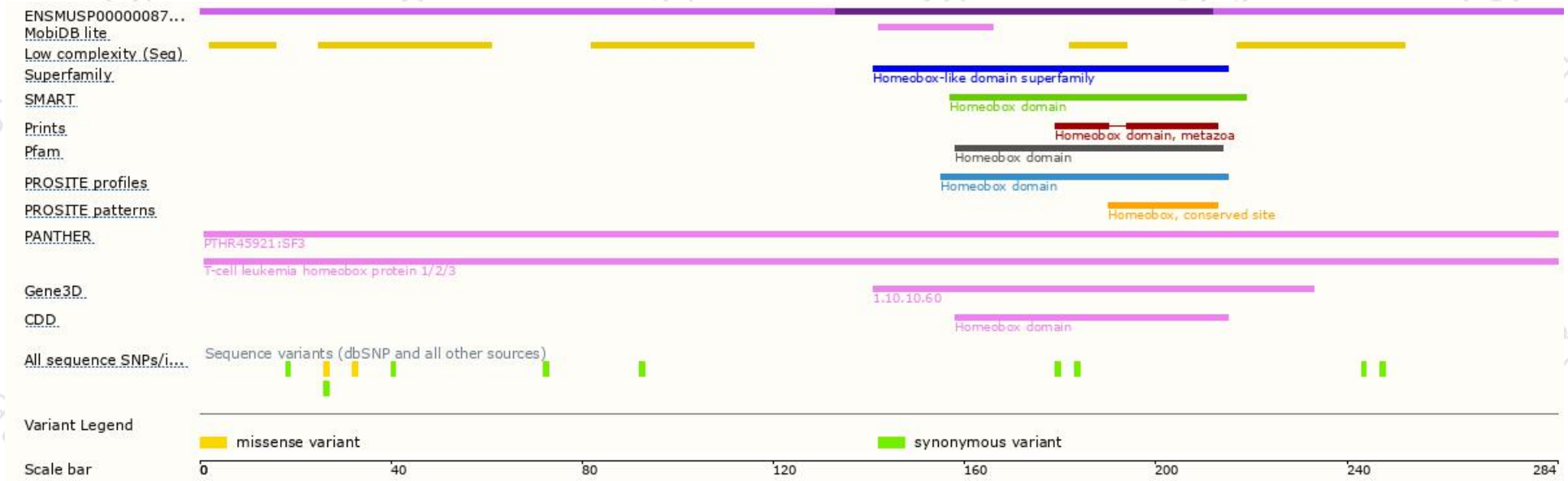
The strategy is based on the design of *Tlx2-201* transcript,the transcription is shown below:



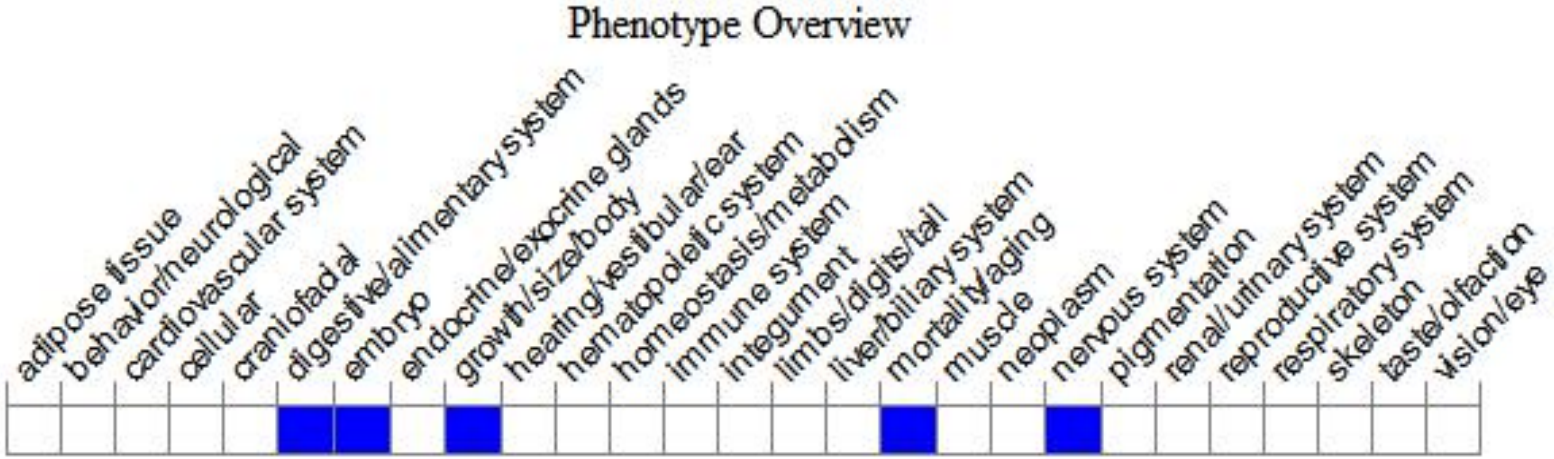
Genomic location distribution



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/).

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

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