Wars2 Cas9-KO *Strategy*

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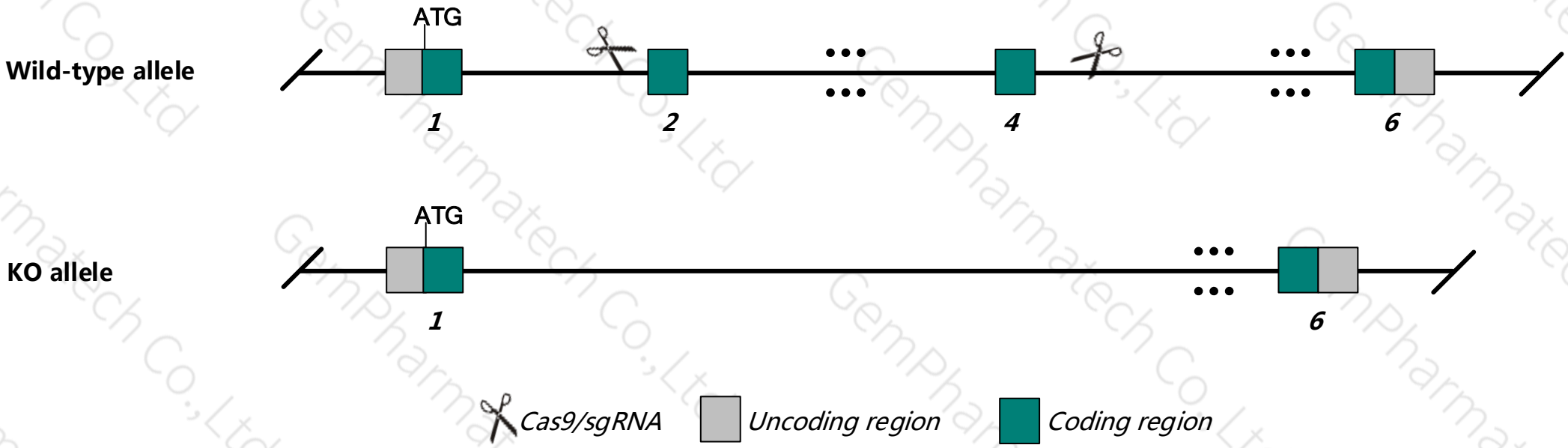
Project Overview

Project Name	***Wars2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Wars2* gene. The schematic diagram is as follows:



Technical routes

- The *Wars2* gene has 5 transcripts.According to the structure of *Wars2* gene, exon2-exon4 of *Wars2*-201 (ENSMUST00000004343.6) transcript is recommended as the knockout region.The region contains 425bp coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Wars2* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9, sgRNA 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，Mice homozygous for an ENU-induced mutation exhibit decreased body fat mass, cardiomyopathy, and progressive hearing loss.
- The insertion position of the 3rd loxp is close to the 5th end of the Gm42717 gene, and the insertion of loxp may affect the regulation of the 5th end of the Gm42717 gene.
- The *Wars2* gene is located on the Chr3. 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）

Wars2 tryptophanyl tRNA synthetase 2 (mitochondrial) [*Mus musculus* (house mouse)]

Gene ID: 70560, updated on 9-Sep-2018

Summary

Official Symbol	Wars2 provided by MGI
Official Full Name	tryptophanyl tRNA synthetase 2 (mitochondrial) provided by MGI
Primary source	MGI:MGI:1917810
See related	Ensembl:ENSMUSG00000004233 Vega:OTTMUSG00000006862
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	TrpRS; AI413375; 5730427B17Rik; 9430020O07Rik
Expression	Ubiquitous expression in limb E14.5 (RPKM 1.9), CNS E11.5 (RPKM 1.5) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 5 transcripts, and all transcripts are shown below:

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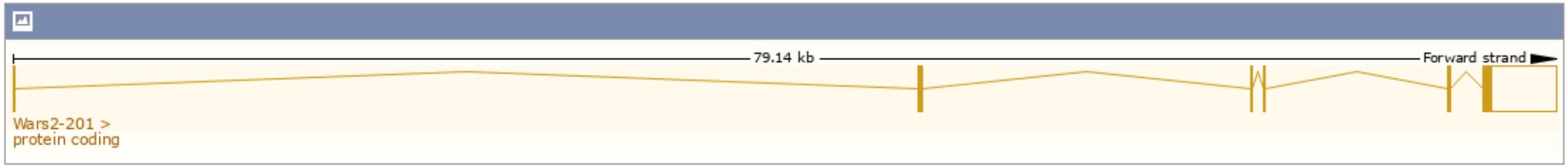
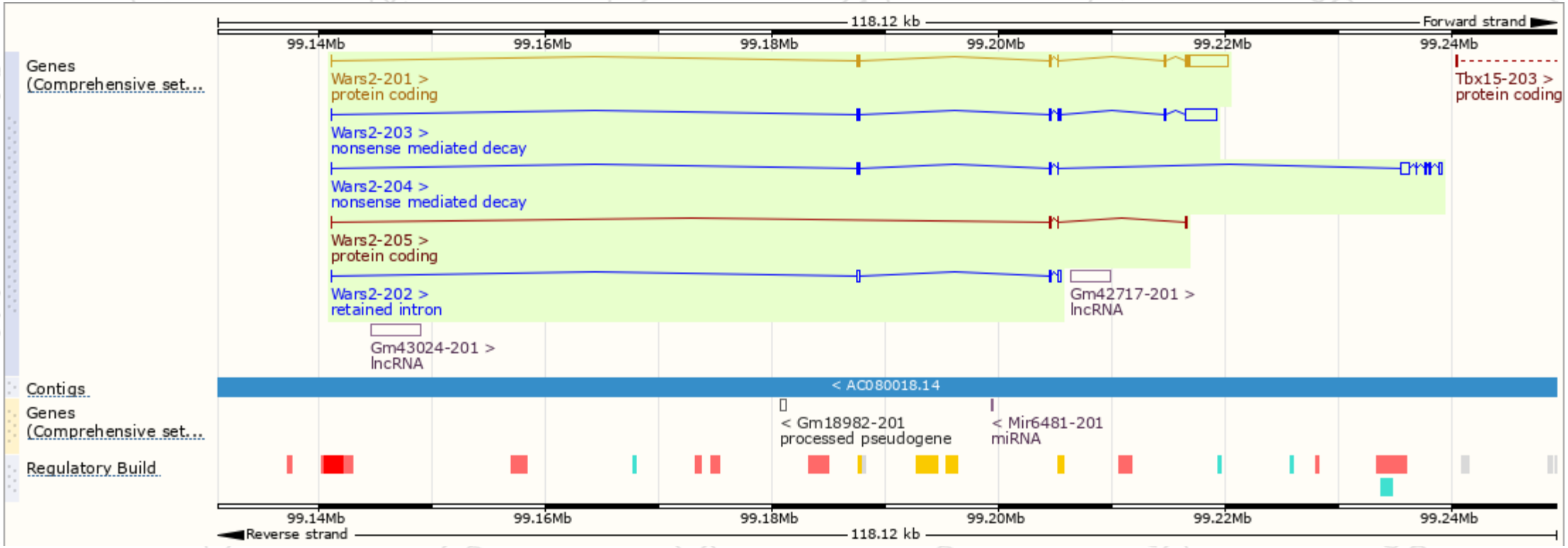
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Wars2-201	ENSMUST00000004343.6	4410	360aa	Protein coding	CCDS17672	Q9CYK1	NM_027462 NP_081738	TSL:1 GENCODE basic APPRIS P1
Wars2-205	ENSMUST00000198044.1	403	106aa	Protein coding	-	A0A0G2JDG7	-	TSL:3 GENCODE basic
Wars2-203	ENSMUST00000135960.7	3457	175aa	Nonsense mediated decay	-	A0A0G2JDR5	-	TSL:1
Wars2-204	ENSMUST00000145650.7	1840	188aa	Nonsense mediated decay	-	Q8BZQ9	-	TSL:1
Wars2-202	ENSMUST00000126875.1	666	No protein	Retained intron	-	-	-	TSL:3

The strategy is based on the design of *Wars2*-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,Homozygous mutant mice are viable and fertile, but differ from wild-type with respect to interleukin 12 mediated T cell function.

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

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