Ctc1 Cas9-KO Strategy

Designer: Yun Li

Reviewer: Longyun Hu

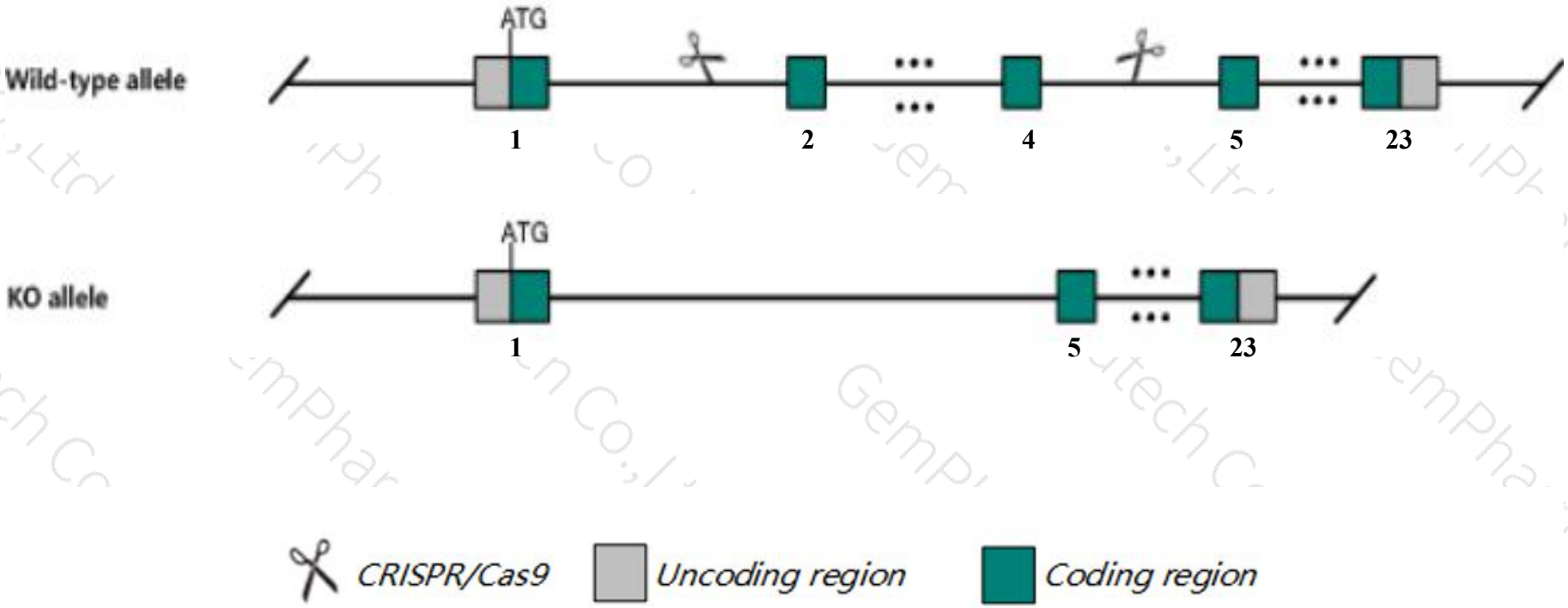
Design Date: 2020-7-31

Project Overview

Project Name	***Ctc1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ctc1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Ctc1* gene has 12 transcripts. According to the structure of *Ctc1* gene, exon2-exon4 of *Ctc1-201*(ENSMUST00000021278.13) transcript is recommended as the knockout region.The region contains 614bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Ctc1* 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,mice homozygous for a targeted allele exhibit defective telomere replication that leads to stem cell exhaustion, bone marrow failure and premature death.

➢ The *Ctc1* gene is located on the Chr11. 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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Ctc1 CTS telomere maintenance complex component 1 [Mus musculus (house mouse)]

Gene ID: 68964, updated on 13-Mar-2020

Summary

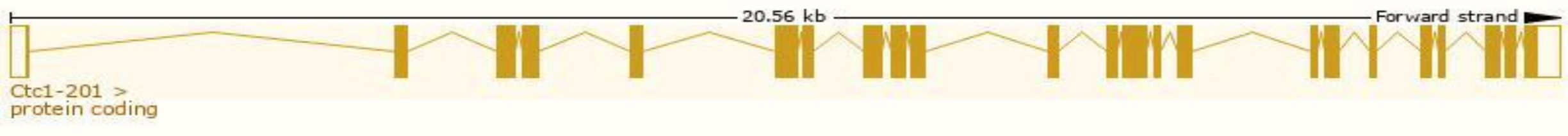
Official Symbol	Ctc1 provided by MGI
Official Full Name	CTS telomere maintenance complex component 1 provided byMGI
Primary source	MGI:MGI:1916214
See related	Ensembl:ENSMUSG00000020898
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1500010J02Rik, AAF-132, AAF132
Expression	Ubiquitous expression in whole brain E14.5 (RPKM 26.5), CNS E14 (RPKM 22.6) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

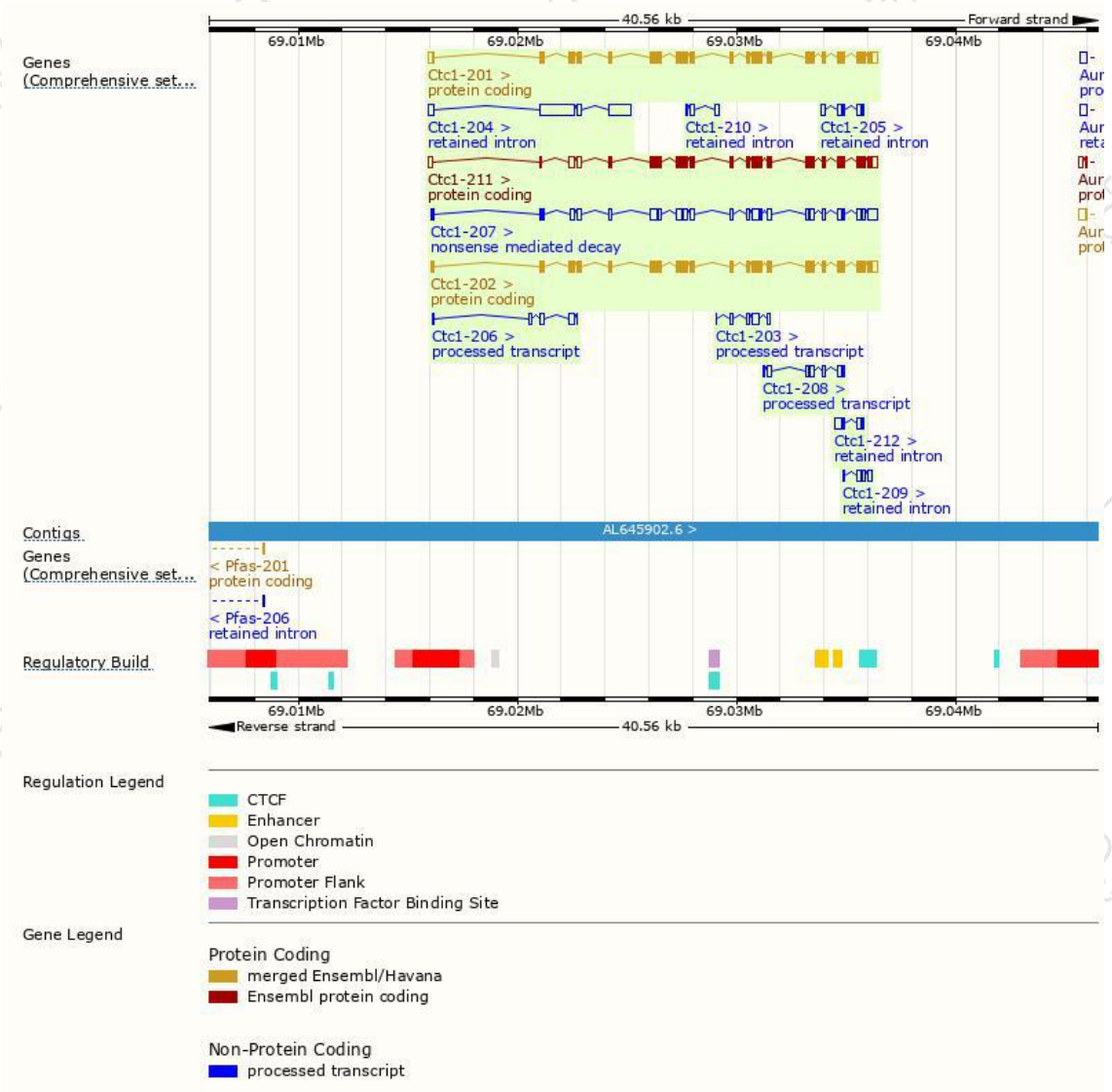
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ctc1-201	ENSMUST00000021278.13	4164	1211aa	Protein coding	CCDS48824	Q5SUQ9	TSL:1 GENCODE basic APPRIS ALT2
Ctc1-211	ENSMUST00000161455.7	3989	965aa	Protein coding	CCDS70214	E0CXE7	TSL:2 GENCODE basic APPRIS ALT2
Ctc1-202	ENSMUST00000116359.2	3978	1212aa	Protein coding	CCDS36189	Q5SUQ9	TSL:1 GENCODE basic APPRIS P3
Ctc1-207	ENSMUST00000152979.7	3976	67aa	Nonsense mediated decay	-	E0CXT1	TSL:1
Ctc1-208	ENSMUST00000154126.7	823	No protein	Processed transcript	-	-	TSL:5
Ctc1-203	ENSMUST00000123871.1	752	No protein	Processed transcript	-	-	TSL:3
Ctc1-206	ENSMUST00000146621.2	645	No protein	Processed transcript	-	-	TSL:3
Ctc1-204	ENSMUST00000135611.7	3035	No protein	Retained intron	-	-	TSL:1
Ctc1-205	ENSMUST00000143404.7	593	No protein	Retained intron	-	-	TSL:3
Ctc1-212	ENSMUST00000162256.7	530	No protein	Retained intron	-	-	TSL:3
Ctc1-209	ENSMUST00000161271.1	516	No protein	Retained intron	-	-	TSL:3
Ctc1-210	ENSMUST00000161384.1	466	No protein	Retained intron	-	-	TSL:3

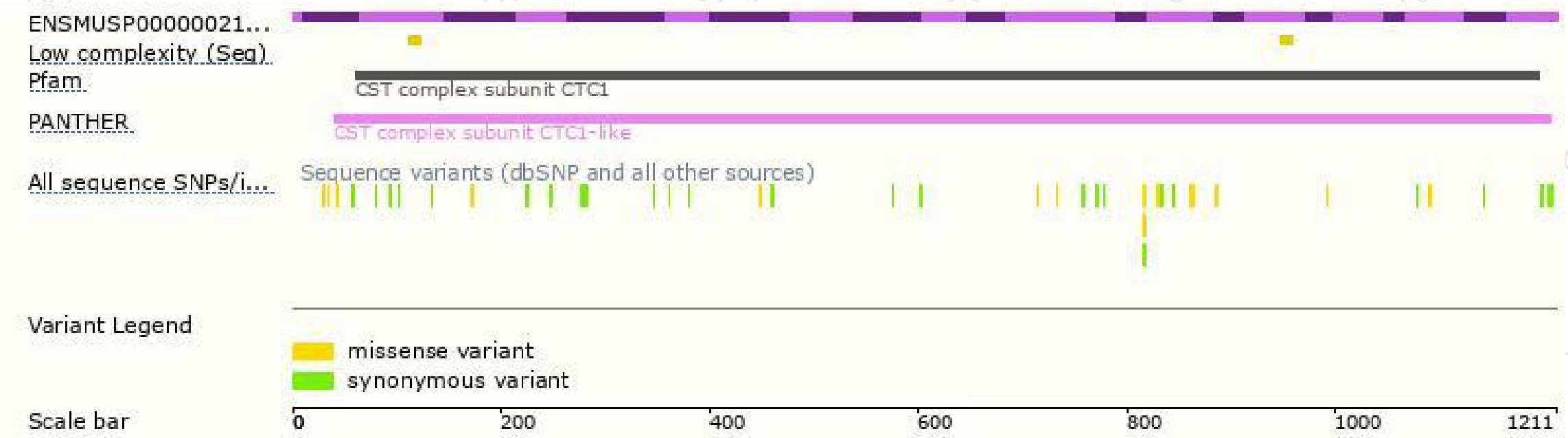
The strategy is based on the design of *Ctc1-201* transcript,the transcription is shown below:



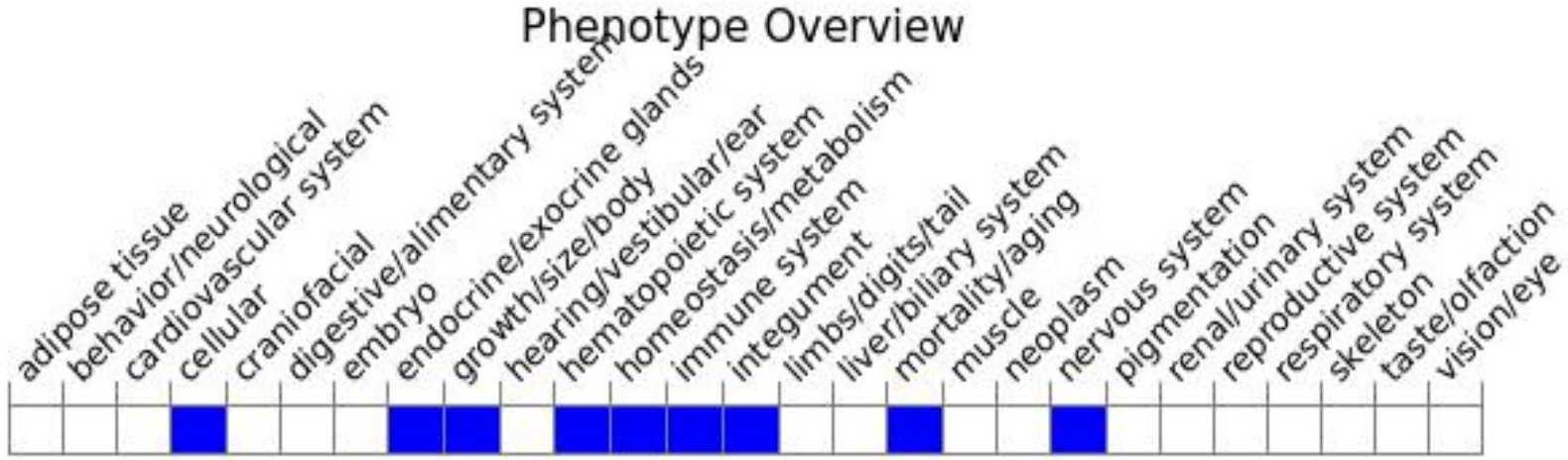
Genomic location distribution



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 targeted allele exhibit defective telomere replication that leads to stem cell exhaustion, bone marrow failure and premature death.

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

Tel: 400-9660890

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GemPharmatech Co.,Ltd