Thoc1 Cas9-CKO Strategy

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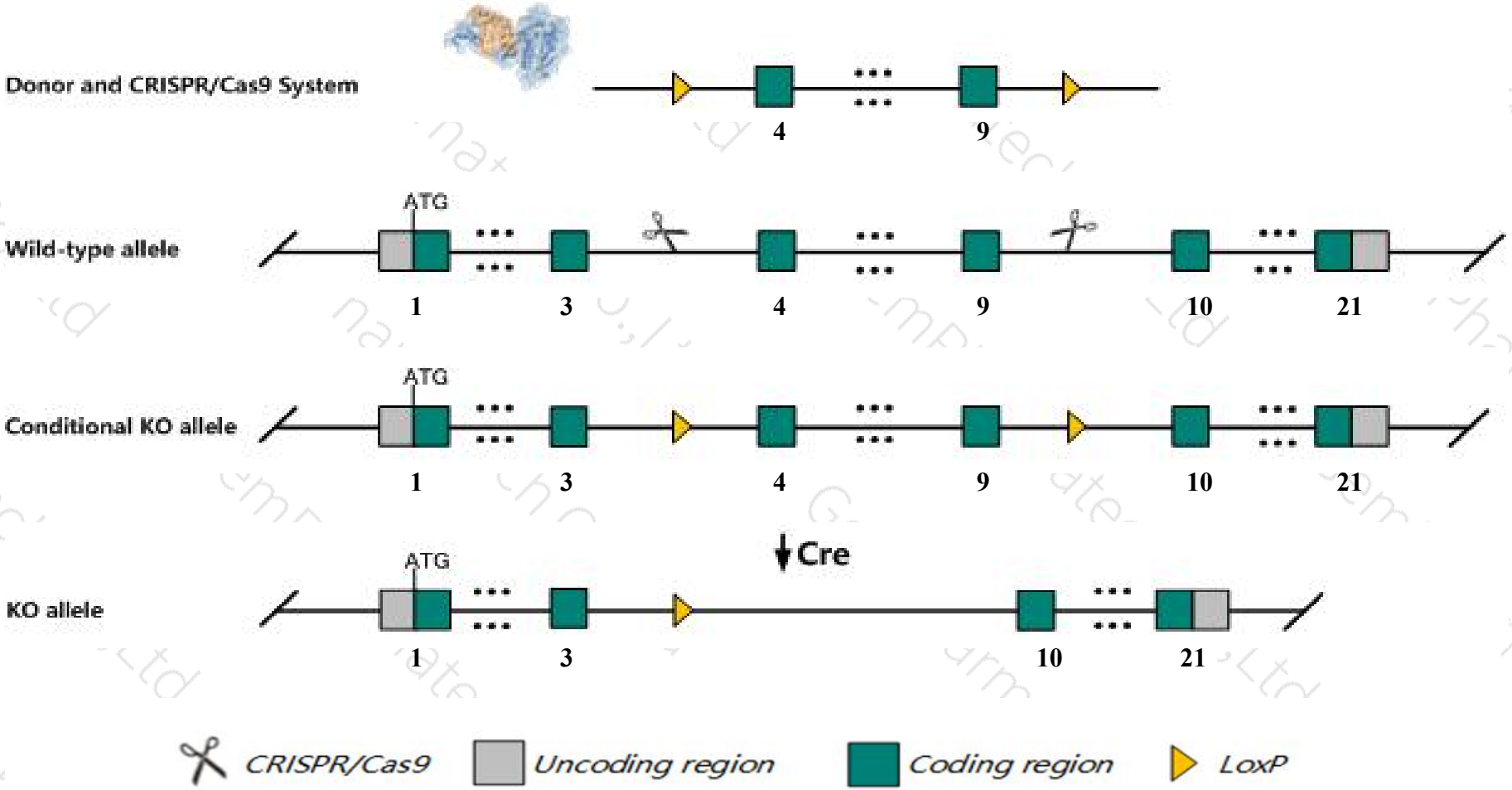
Project Overview

Project Name	***Thoc1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Thoc1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Thoc1* gene has 13 transcripts. According to the structure of *Thoc1* gene, exon4-exon9 of *Thoc1-201*(ENSMUST00000025137.8) transcript is recommended as the knockout region.The region contains 488bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Thoc1* gene. The brief process is as follows:CRISPR/Cas9 system 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.

Notice

➢ According to the existing MGI data,mutations in this gene result in embryonic lethality around implantation in homozygotes.

➢ The *Thoc1* gene is located on the Chr18. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Thoc1 THO complex 1 [*Mus musculus* (house mouse)]

Gene ID: 225160, updated on 26-Jun-2020

Summary

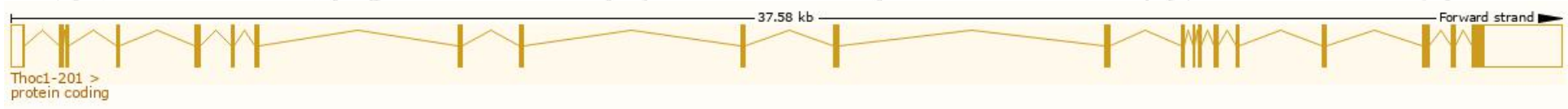
Official Symbol	Thoc1 provided by MGI
Official Full Name	THO complex 1 provided by MGI
Primary source	MGI:MGI:1919668
See related	Ensembl:ENSMUSG00000024287
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	NMP-84; AW107452; 3110002N20Rik
Expression	Broad expression in CNS E11.5 (RPKM 14.3), CNS E18 (RPKM 13.3) and 19 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

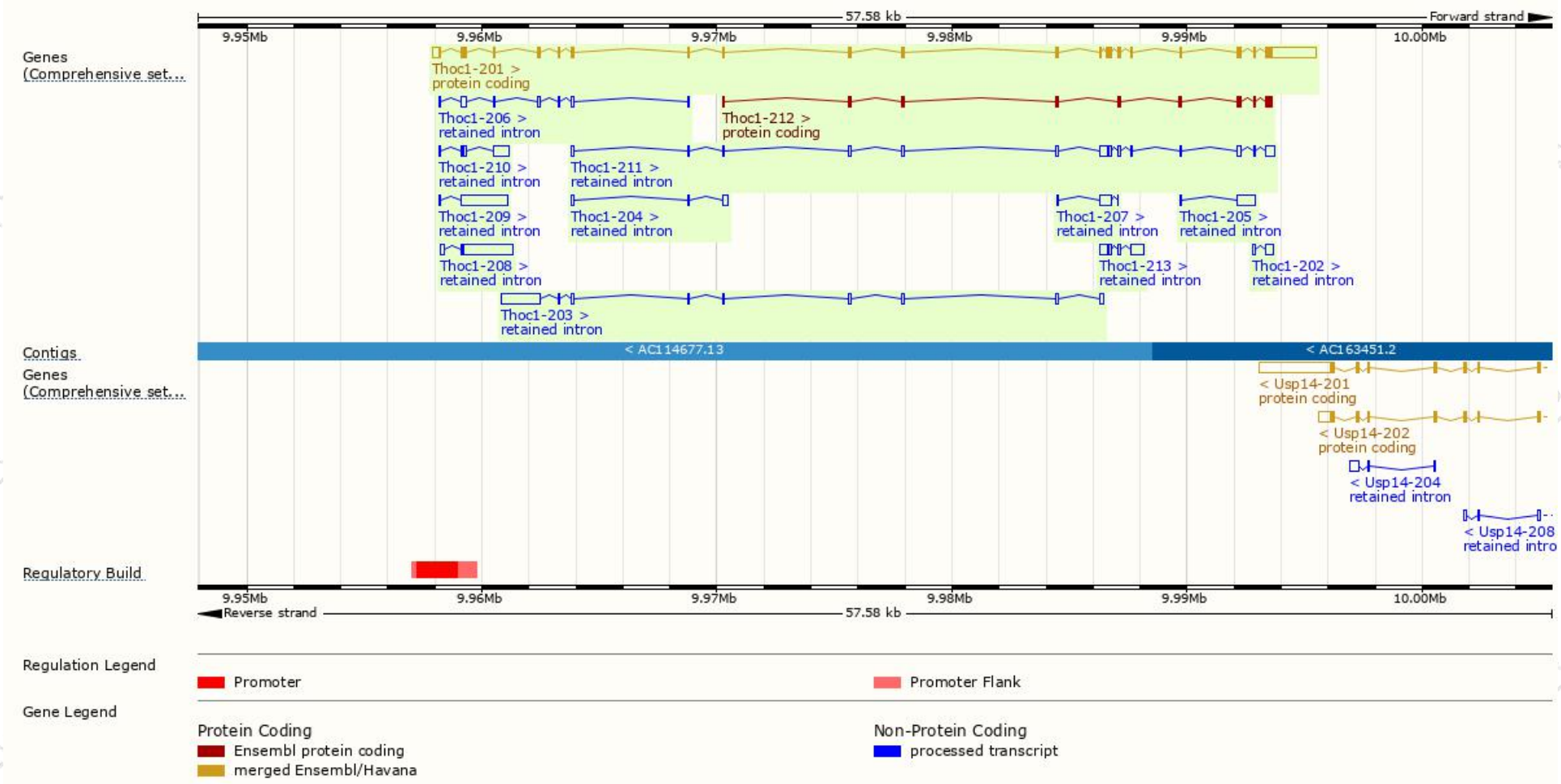
The gene has 13 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein ▲	Biotype	CCDS	UniProt	Flags
Thoc1-212	ENSMUST00000234975.1	1158	385aa	Protein coding	-	A0A3Q4EBV8	CDS 5' incomplete
Thoc1-201	ENSMUST00000025137.8	4134	657aa	Protein coding	CCDS37733	Q8R3N6	TSL:1 GENCODE basic APPRIS P1
Thoc1-203	ENSMUST00000234324.1	2437	No protein	Retained intron	-	-	-
Thoc1-208	ENSMUST00000234622.1	2303	No protein	Retained intron	-	-	-
Thoc1-209	ENSMUST00000234805.1	2047	No protein	Retained intron	-	-	-
Thoc1-211	ENSMUST00000234901.1	1875	No protein	Retained intron	-	-	-
Thoc1-213	ENSMUST00000235035.1	1039	No protein	Retained intron	-	-	-
Thoc1-210	ENSMUST00000234890.1	891	No protein	Retained intron	-	-	-
Thoc1-205	ENSMUST00000234370.1	851	No protein	Retained intron	-	-	-
Thoc1-206	ENSMUST00000234423.1	680	No protein	Retained intron	-	-	-
Thoc1-207	ENSMUST00000234600.1	547	No protein	Retained intron	-	-	-
Thoc1-202	ENSMUST00000234111.1	500	No protein	Retained intron	-	-	-
Thoc1-204	ENSMUST00000234330.1	412	No protein	Retained intron	-	-	-

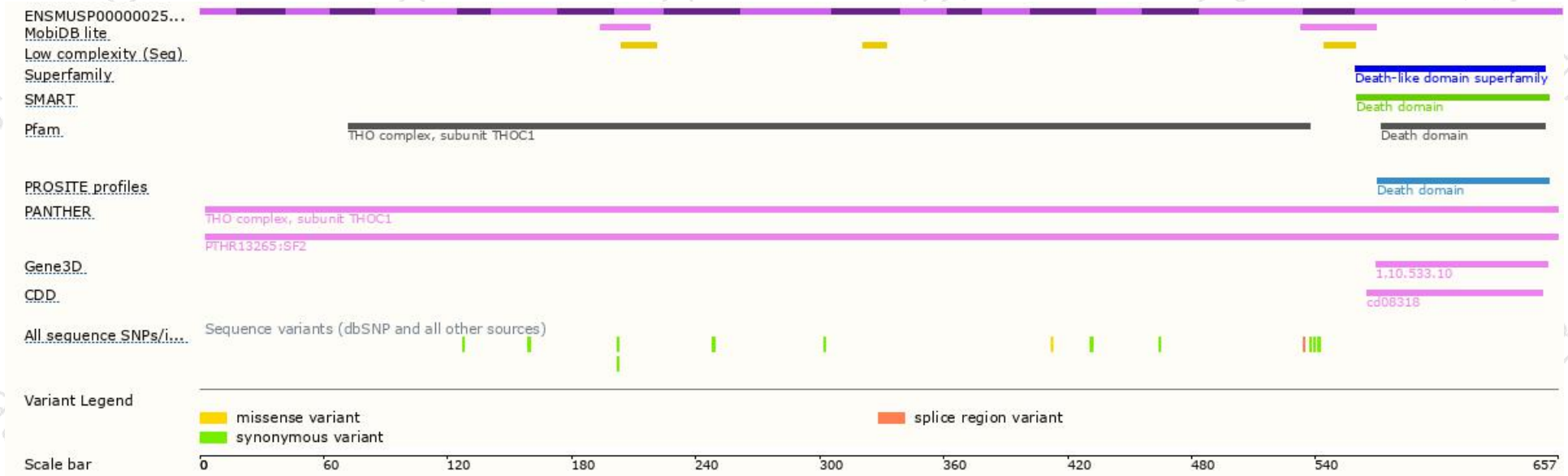
The strategy is based on the design of *Thoc1-201* transcript,the transcription is shown below:



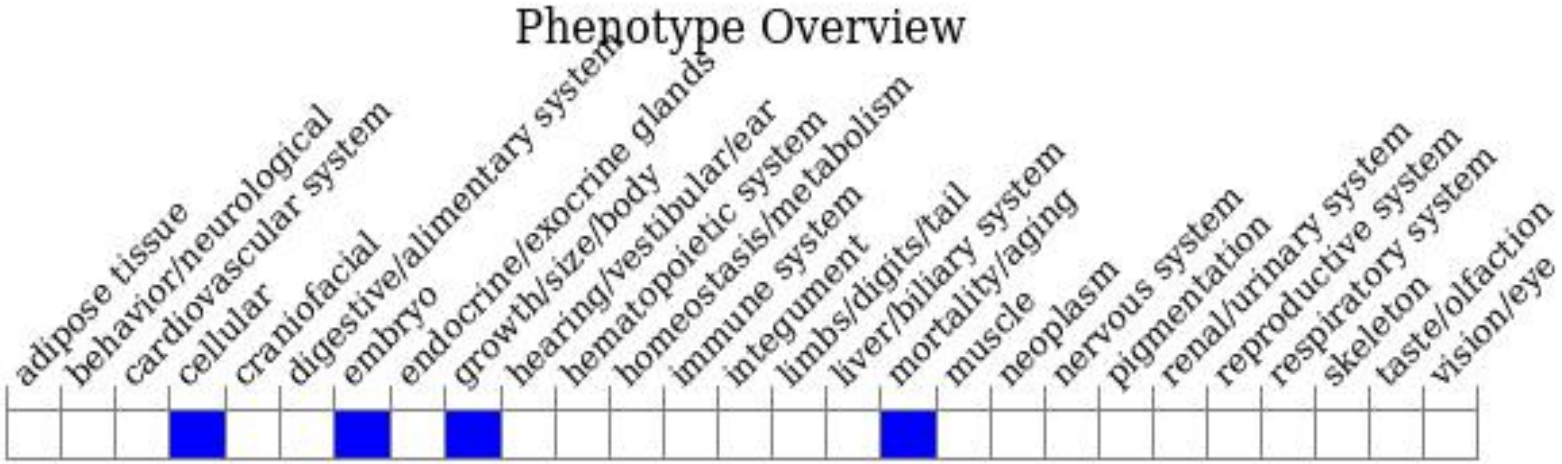
Genomic location distribution



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,mutations in this gene result in embryonic lethality around implantation in homozygotes.

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

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