

Ttll3 Cas9-CKO Strategy

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Project Overview



Project Name

Ttll3

Project type

Cas9-CKO

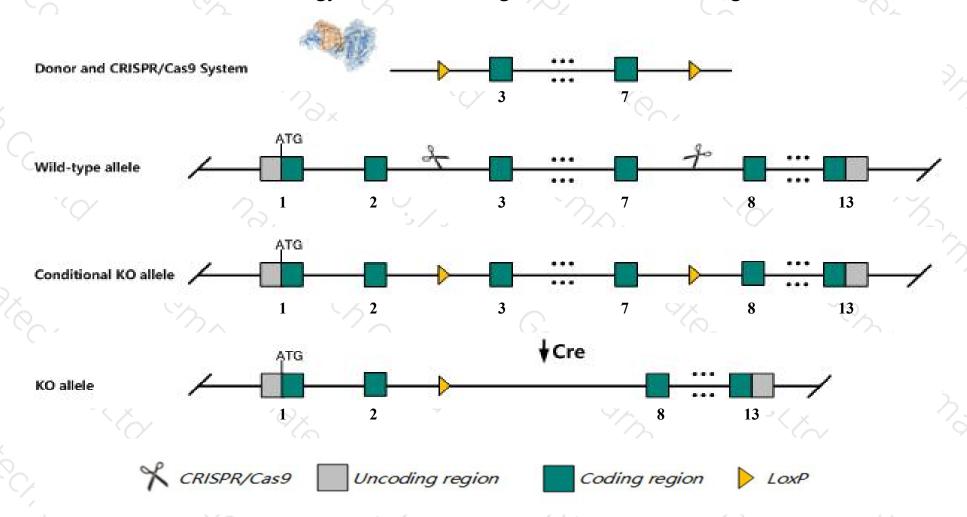
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ttll3* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Ttll3* gene has 9 transcripts. According to the structure of *Ttll3* gene, exon3-exon7 of *Ttll3-201*(ENSMUST00000032414.10) transcript is recommended as the knockout region. The region contains 616bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Ttll3* 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, mice homozygous for a knock-out allele exhibit a reduced number of primary cilia in colon epithelia accompanied by an increased rate of cell division which is compensated by faster tissue turnover in the colon. mice exhibit increased incidence of colon tumors by chemical induction. retinal photoreceptor connecting cilia are also affected, resulting in progressive degeneration of the photoreceptors.
- > The *Ttll3* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)



Ttll3 tubulin tyrosine ligase-like family, member 3 [Mus musculus (house mouse)]

Gene ID: 101100, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Ttll3 provided by MGI

Official Full Name tubulin tyrosine ligase-like family, member 3 provided by MGI

Primary source MGI:MGI:2141418

See related Ensembl: ENSMUSG00000030276

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 4833441J24Rik, Al450050

Expression Ubiquitous expression in testis adult (RPKM 32.1), thymus adult (RPKM 12.4) and 28 other tissues See more

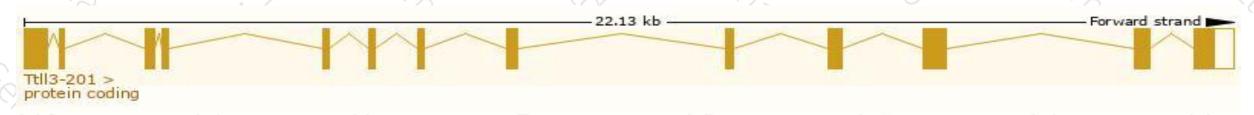
Transcript information (Ensembl)



The gene has 9 transcripts, all transcripts are shown below:

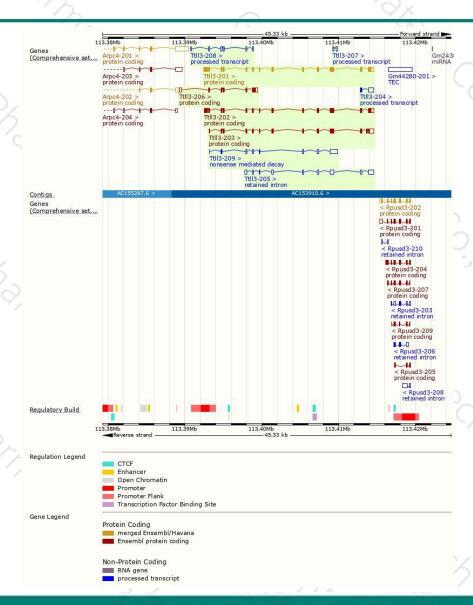
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ttll3-201	ENSMUST00000032414.10	3168	927aa	Protein coding	CCDS39593	A4Q9E5	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P2
Ttll3-206	ENSMUST00000204026.2	1472	<u>287aa</u>	Protein coding	CCDS85118	A4Q9E5	TSL:1 GENCODE basic
Ttll3-202	ENSMUST00000038889.11	3114	928aa	Protein coding	1/2	H7BX03	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT2
Ttll3-203	ENSMUST00000203524.2	2745	<u>721aa</u>	Protein coding	12	F8VQA1	CDS 5' incomplete TSL:5
Ttll3-209	ENSMUST00000205017.2	1886	<u>68aa</u>	Nonsense mediated decay	15	F6T422	CDS 5' incomplete TSL:1
Ttll3-204	ENSMUST00000203880.1	827	No protein	Processed transcript	, 8 8		TSL:3
Ttll3-208	ENSMUST00000204683.1	782	No protein	Processed transcript	1/4	-	TSL:5
Ttll3-207	ENSMUST00000204255.1	396	No protein	Processed transcript	14	20	TSL:3
Ttll3-205	ENSMUST00000203925.1	2517	No protein	Retained intron	65	-	TSL:1

The strategy is based on the design of *Ttll3-201* transcript, the transcription is shown below:



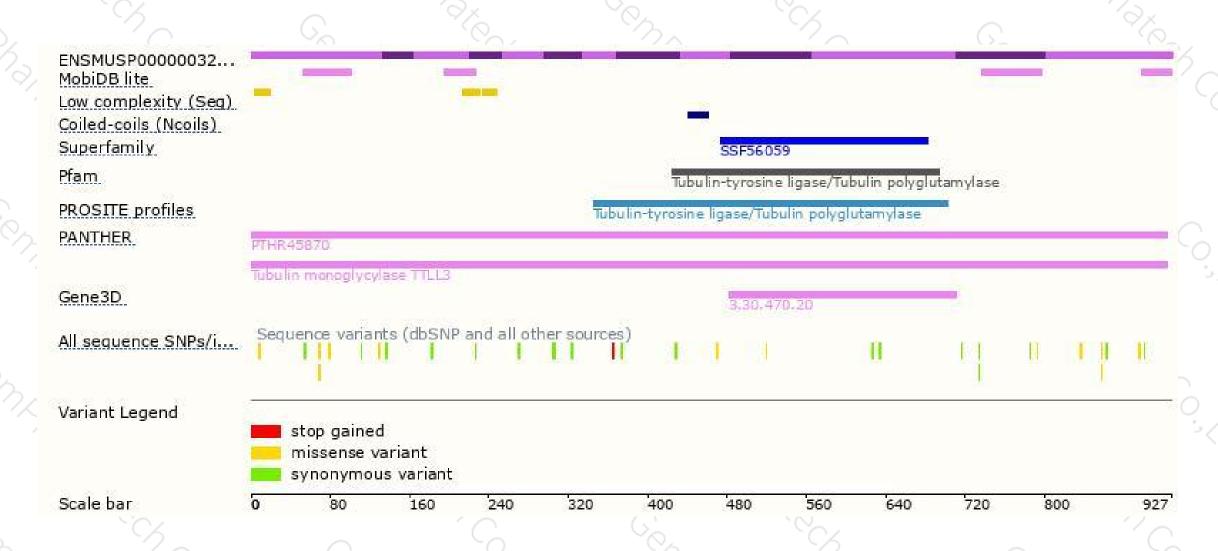
Genomic location distribution





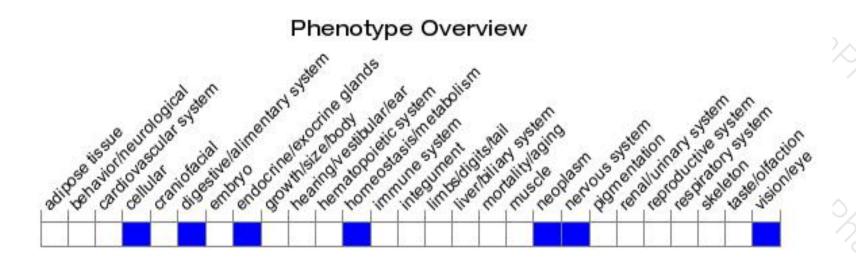
Protein domain





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 knock-out allele exhibit a reduced number of primary cilia in colon epithelia accompanied by an increased rate of cell division which is compensated by faster tissue turnover in the colon. Mice exhibit increased incidence of colon tumors by chemical induction. Retinal photoreceptor connecting cilia are also affected, resulting in progressive degeneration of the photoreceptors.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





