

# **Thrap3** Cas9-CKO Strategy

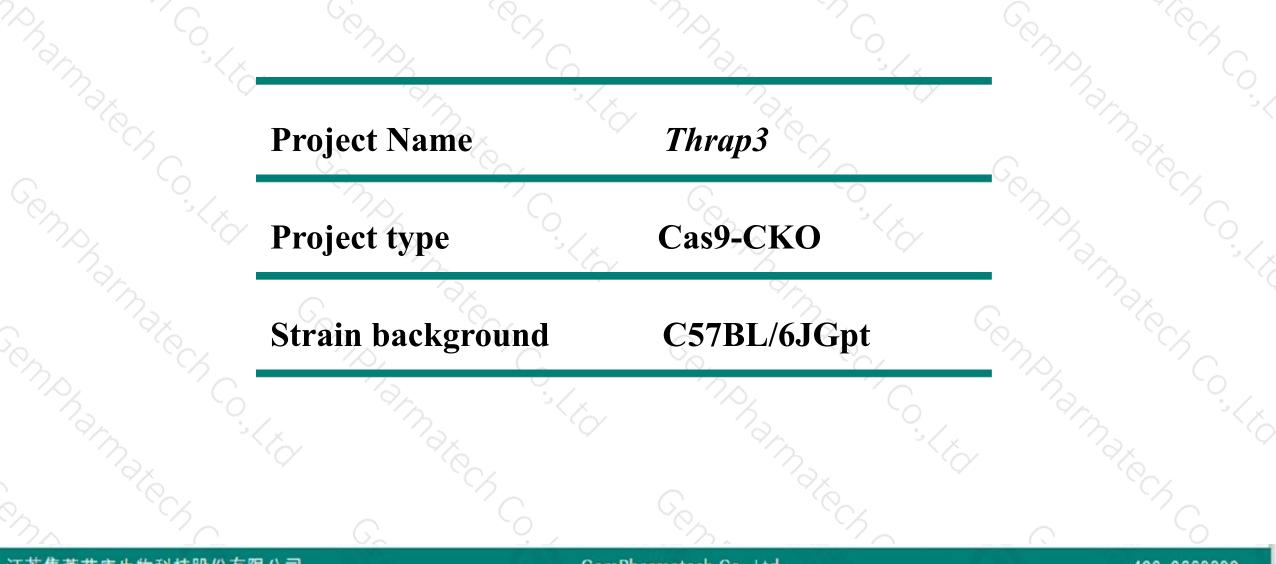
Designer: Reviewer:

**Design Date:** 

Daohua Xu Huimin Su 2020-1-15

### **Project Overview**





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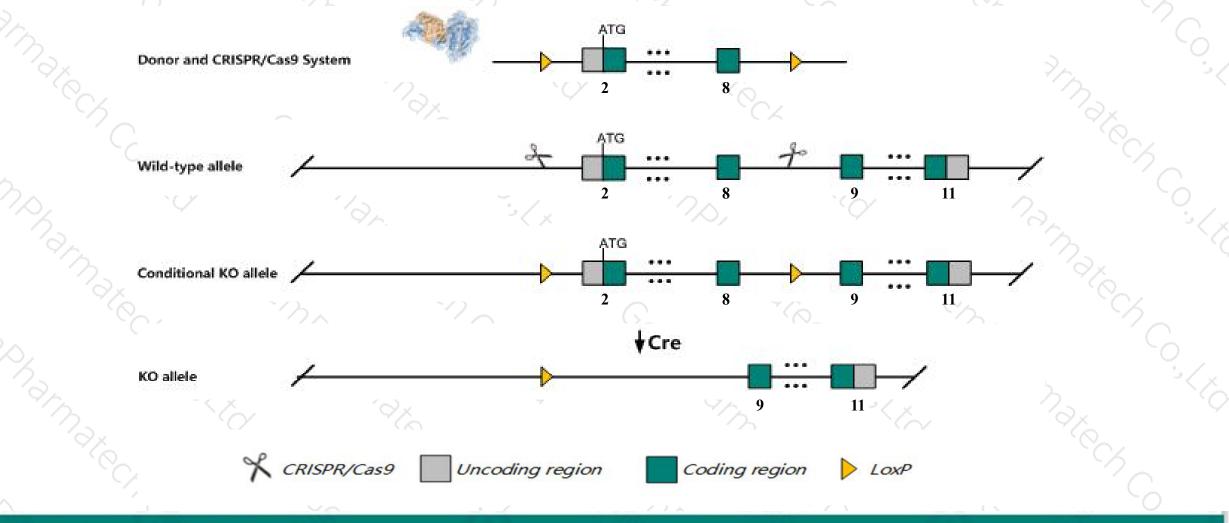
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### **Conditional Knockout strategy**



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This model will use CRISPR/Cas9 technology to edit the *Thrap3* gene. The schematic diagram is as follows:



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The *Thrap3* gene has 18 transcripts. According to the structure of *Thrap3* gene, exon2-exon8 of *Thrap3-201* (ENSMUST0000080919.11) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Thrap3* 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.



- The *Thrap3* gene is located on the Chr4. 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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#### Thrap3 thyroid hormone receptor associated protein 3 [Mus musculus (house mouse)]

Gene ID: 230753, updated on 3-Feb-2019

#### Summary

Official Symbol	Thrap3 provided by MGI
Official Full Name	thyroid hormone receptor associated protein 3 provided by MGI
Primary source	MGI:MGI:2442637
See related	Ensembl:ENSMUSG0000043962
Gene type	protein coding
<b>RefSeq status</b>	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	9330151F09Rik, B230333E16Rik, Trap150
Expression	Ubiquitous expression in CNS E11.5 (RPKM 45.1), limb E14.5 (RPKM 31.5) and 28 other tissues See more
Orthologs	human all

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### **Transcript information (Ensembl)**



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#### The gene has 18 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Thrap3-201	ENSMUST0000080919.11	4378	<u>951aa</u>	Protein coding	CCDS18645	<u>Q569Z6</u>	TSL:1 GENCODE basic APPRIS P1
Thrap3-203	ENSMUST00000106142.7	2744	<u>709aa</u>	Protein coding		Q8BZN7	TSL:1 GENCODE basic
Thrap3-210	ENSMUST00000142125.1	673	<u>27aa</u>	Protein coding	3 <b>-</b>	A2AJI7	CDS 3' incomplete TSL:3
Thrap3-215	ENSMUST00000163459.7	583	<u>194aa</u>	Protein coding	1 C.	<u>F6YH92</u>	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Thrap3-214	ENSMUST00000163306.1	407	<u>135aa</u>	Protein coding		F6YSQ2	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Thrap3-206	ENSMUST00000130334.1	378	<u>29aa</u>	Protein coding	8-	A2AJI6	CDS 3' incomplete TSL:3
Thrap3-202	ENSMUST00000106141.2	361	<u>25aa</u>	Protein coding	3 <del>.</del>	E9Q6C5	CDS 3' incomplete TSL:3
Thrap3-208	ENSMUST00000136157.7	350	<u>53aa</u>	Protein coding	<u>.</u>	A2AJI3	CDS 3' incomplete TSL:5
Thrap3-217	ENSMUST00000169403.7	339	<u>50aa</u>	Protein coding	17	E9Q5X3	CDS 3' incomplete TSL:3
Thrap3-204	ENSMUST00000123008.7	693	<u>59aa</u>	Nonsense mediated decay		E9Q494	TSL:3
Thrap3-213	ENSMUST00000163176.1	688	<u>43aa</u>	Nonsense mediated decay	3 <b>-</b>	F7AB07	CDS 5' incomplete TSL:3
Thrap3-211	ENSMUST00000151728.1	2238	No protein	Retained intron	<u>.</u>		TSL1
Thrap3-207	ENSMUST00000130477.1	578	No protein	Retained intron	10		TSL:2
Thrap3-216	ENSMUST00000168568.7	521	No protein	Retained intron	8-		TSL:2
Thrap3-209	ENSMUST00000140390.1	2656	No protein	IncRNA	-	-	TSL:1
Thrap3-212	ENSMUST00000154702.1	609	No protein	IncRNA	1		TSL:2
Thrap3-205	ENSMUST00000124327.1	590	No protein	IncRNA	10		TSL:2
Thrap3-218	ENSMUST00000169855.1	450	No protein	IncRNA	8-		TSL:3
		1.1		/ /			

The strategy is based on the design of Thrap3-201 transcript, The transcription is shown below

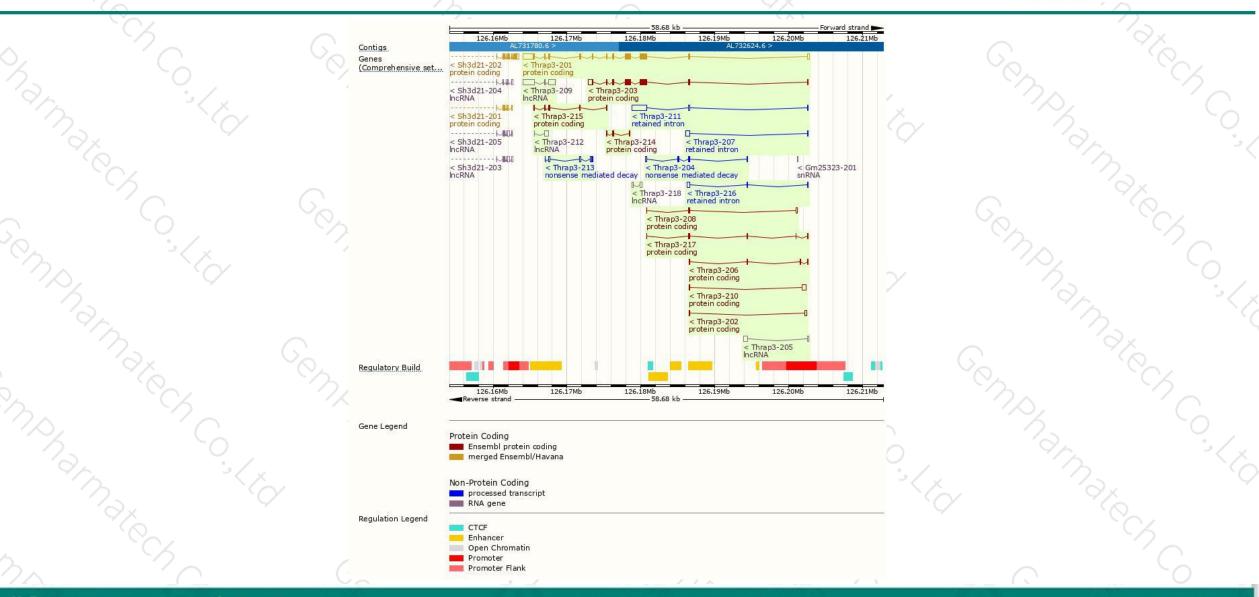
< Thrap3-201 protein coding

Reverse strand -

- 38.68 kb -

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### **Genomic location distribution**



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### **Protein domain**

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24	Variant Legend Scale bar		issense v /nonymo 80	variant us variant 160	240	320	400	480	560	640	720	800	951
	Variant Legend												
	All sequence SNPs/i	1000		receptor-as: nts (dbSNP			es)		1	$\mathbf{I}(\mathbf{I})$	Ĵ.	(1)	0000
	MobiDB lite Low complexity (Seg) Pfam PANTHER		/BCLAF1										
	ENSMUSP00000079 MobiDB lite			5,		ò, ,		3 m					



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



