

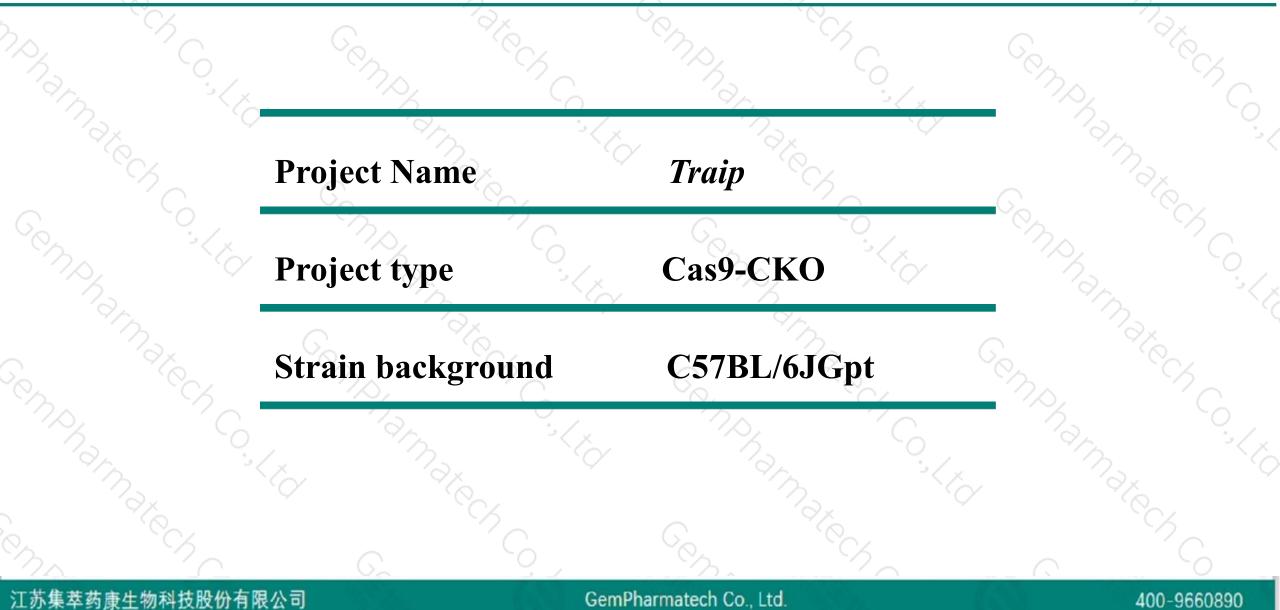
Traip Cas9-CKO Strategy Romphamater Control

Comphannated Co Designer: Shilei Zhu Semphamatech Co

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Project Overview

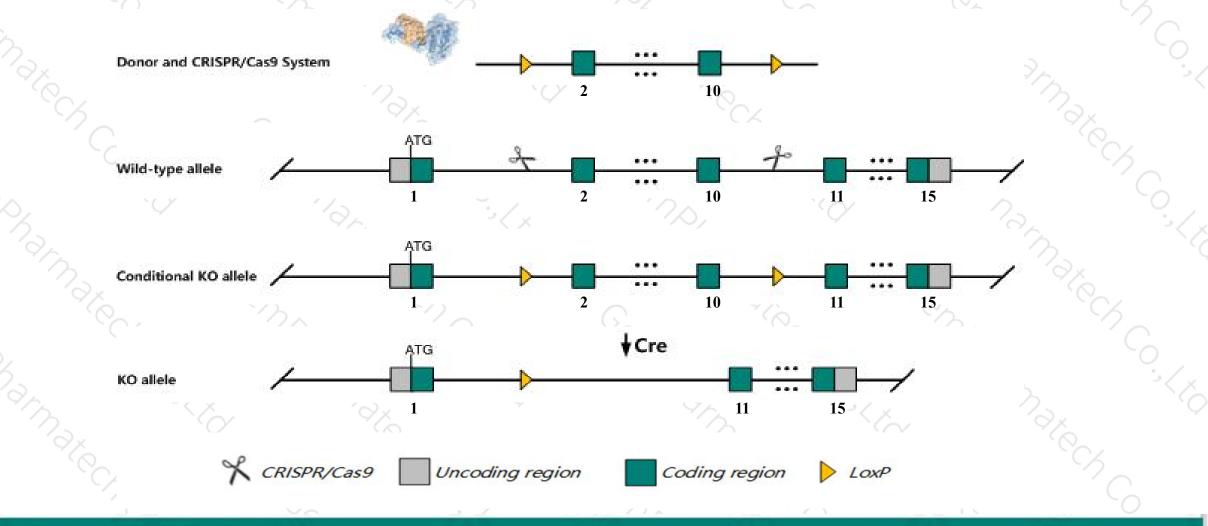




Conditional Knockout strategy



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



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The *Traip* gene has 7 transcripts. According to the structure of *Traip* gene, exon2-exon10 of *Traip-201* (ENSMUST00000049348.8) transcript is recommended as the knockout region. The region contains 786bp coding sequence. Knock out the region will result in disruption of protein function.

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



- According to the existing MGI data, Mice homozygous for a null allele exhibit embryonic lethality at prior to E8.5, embryonic growth retardation, decreased embryonic size, decreased cell proliferation and increased apoptosis.
- The *Traip* gene is located on the Chr9. 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)



\$?

Traip TRAF-interacting protein [Mus musculus (house mouse)]

Gene ID: 22036, updated on 5-Feb-2019

Summary

Traip provided by MGI
TRAF-interacting protein provided by MGI
MGI:MGI:1096377
Ensembl:ENSMUSG00000032586
protein coding
VALIDATED
Mus musculus
Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Muroidea; Muridae; Murinae; Mus; Mus
Trip
Broad expression in CNS E11.5 (RPKM 3.7), frontal lobe adult (RPKM 3.7) and 19 other tissues See more
human all

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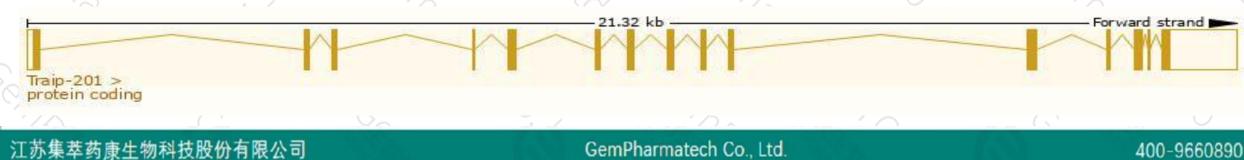
Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

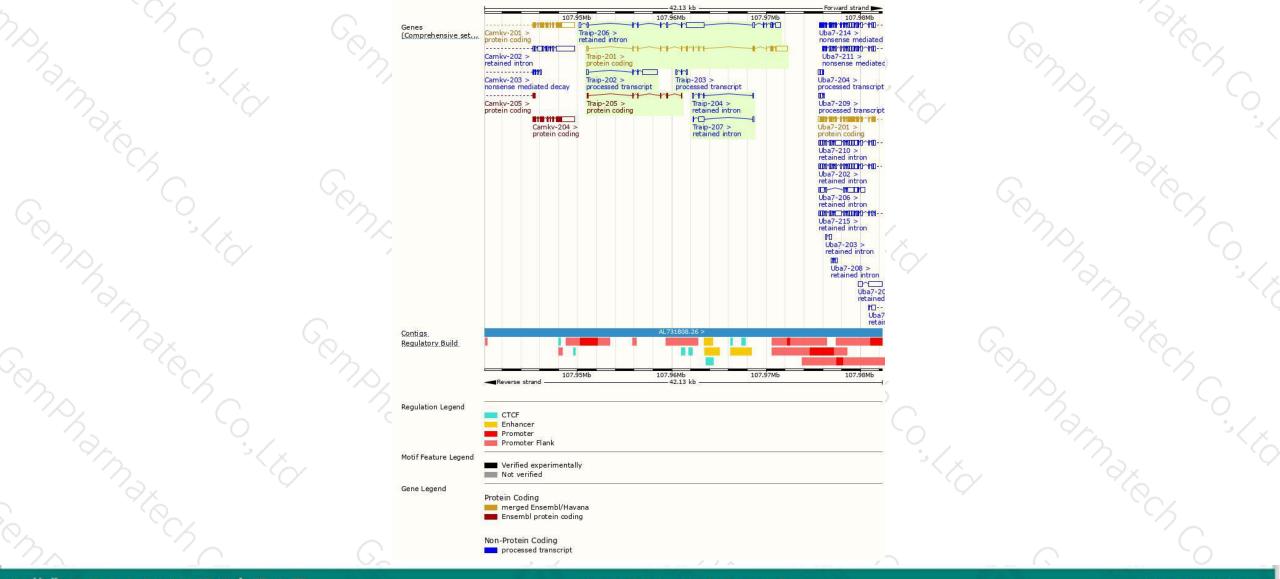
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Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST0000049348.8	2706	<u>470aa</u>	Protein coding	CCDS23511	<u>Q8VIG6</u>	TSL:1 GENCODE basic APPRIS P1
ENSMUST00000194271.1	515	<u>142aa</u>	Protein coding	-	A0A0A6YWT5	CDS 3' incomplete TSL:5
ENSMUST00000192567.1	1949	No protein	Processed transcript	6 2 0	2	TSL:1
ENSMUST00000193715.1	383	No protein	Processed transcript	1926	2	TSL:3
ENSMUST00000194538.5	3680	No protein	Retained intron	187)	54	TSL:2
ENSMUST00000195803.1	844	No protein	Retained intron	-	-	TSL:3
ENSMUST00000194191.1	400	No protein	Retained intron	(2)	-	TSL:3
	ENSMUST0000049348.8 ENSMUST00000194271.1 ENSMUST00000192567.1 ENSMUST00000193715.1 ENSMUST00000194538.5 ENSMUST00000195803.1	ENSMUST0000049348.8 2706 ENSMUST0000194271.1 515 ENSMUST0000192567.1 1949 ENSMUST0000193715.1 383 ENSMUST0000194538.5 3680 ENSMUST0000195803.1 844	ENSMUST0000049348.8 2706 470aa ENSMUST00000194271.1 515 142aa ENSMUST0000192567.1 1949 No protein ENSMUST00000193715.1 383 No protein ENSMUST00000194538.5 3680 No protein	ENSMUST0000049348.82706470aaProtein codingENSMUST0000194271.1515142aaProtein codingENSMUST0000192567.11949No proteinProcessed transcriptENSMUST0000193715.1383No proteinProcessed transcriptENSMUST0000194538.53680No proteinRetained intronENSMUST0000195803.1844No proteinRetained intron	ENSMUST0000049348.82706470aaProtein codingCCDS23511ENSMUST0000194271.1515142aaProtein coding-ENSMUST0000192567.11949No proteinProcessed transcript-ENSMUST0000193715.1383No proteinProcessed transcript-ENSMUST0000194538.53680No proteinRetained intron-ENSMUST0000195803.1844No proteinRetained intron-	ENSMUST000004934882706470aaProtein codingCCDS23511Q8VIG6ENSMUST00001942711515142aaProtein coding-A0A0A6YWT5ENSMUST000019256711949No proteinProcessed transcriptENSMUST00001937151383No proteinProcessed transcriptENSMUST0000194538553680No proteinRetained intronENSMUST00001958031844No proteinRetained intron

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



Genomic location distribution





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Protein domain



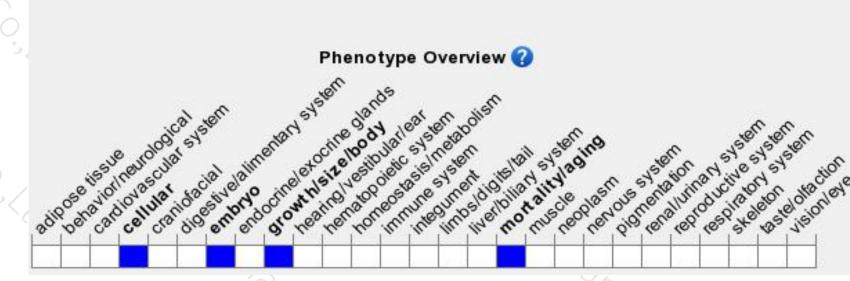
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S.	ENSMUSP00000040 Conserved Domains Coiled-coils (Ncoils) hmmpanther	PTHR22937:SP	52		122			-					
	Superfamily domains	PTHR22937 SSF57850	2	SSF4657	79		-						
2	SMART domains	Zinc finger, R	ING-type										6
	Pfam_domain_	Zinc finger, R	1941										
	PROSITE profiles Gene3D	Zinc finger, R Zinc finger, RI	- Second and the second	D-type									
5,	All sequence SNPs/i	Sequence va	iants (dbSN	VP and all ot	her source	25)	1	1	mí.			1	12
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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 null allele exhibit embryonic lethality at prior to E8.5, embryonic growth retardation, decreased embryonic size, decreased cell proliferation and increased apoptosis.



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



