

Tab2 Cas9-CKO Strategy

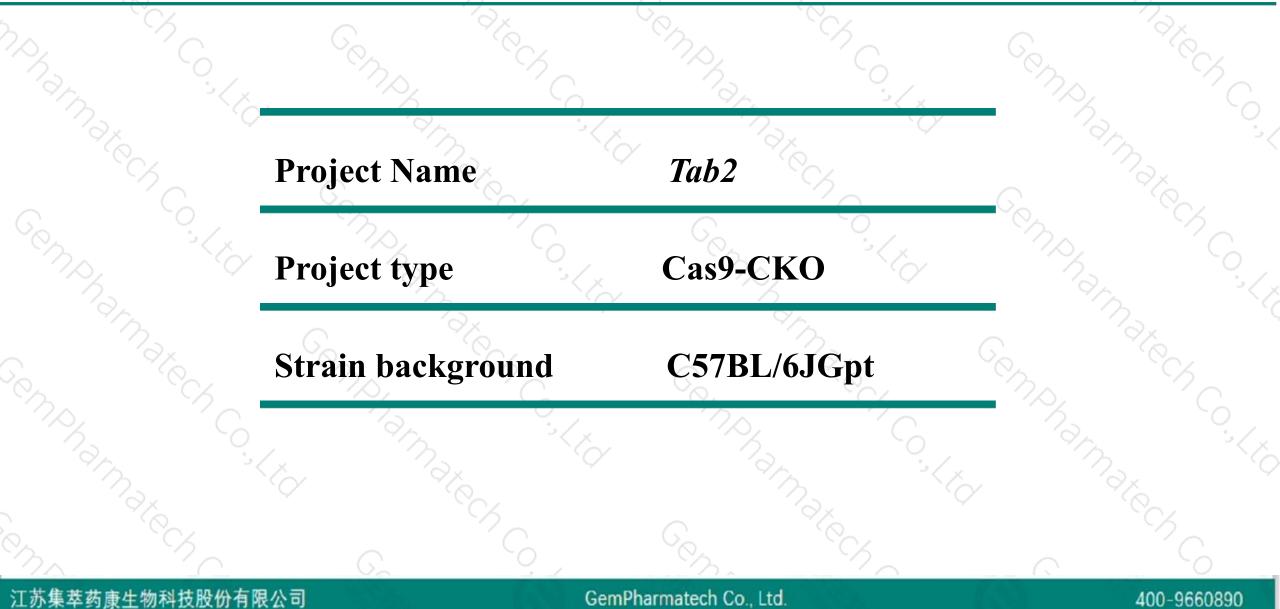
Designer: Reviewer:

Design Date:

Huan Wang Huan Fan 2020-3-6

Project Overview



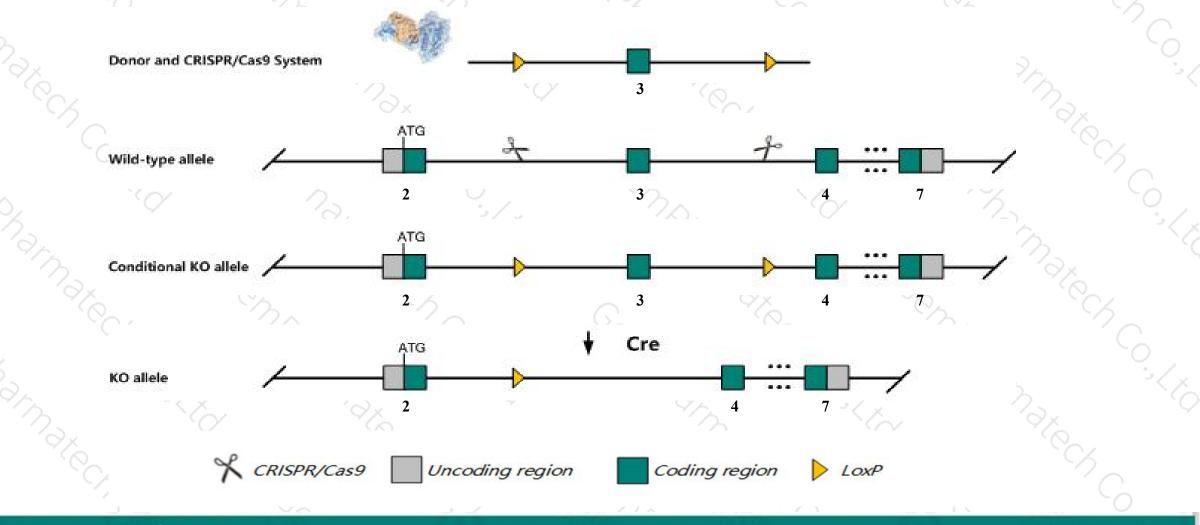


Conditional Knockout strategy



400-9660890

This model will use CRISPR/Cas9 technology to edit the Tab2 gene. The schematic diagram is as follows:



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The *Tab2* gene has 5 transcripts. According to the structure of *Tab2* gene, exon3 of *Tab2-204* (ENSMUST00000146444.7) transcript is recommended as the knockout region. The region contains 1501bp coding sequence.
Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Tab2* 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, Embryos homozygous for a knock-out allele are viable up to E9.5. Embryos homozygous for a different knock-out allele are normal and viable up to E11.5 but become pale and anemic, exhibit liver hemorrhage and increased apoptosis of hepatoblasts, and die by E12.5.
- The Tab2 gene is located on the Chr10. 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)



\$?

Tab2 TGF-beta activated kinase 1/MAP3K7 binding protein 2 [Mus musculus (house mouse)]

Gene ID: 68652, updated on 19-Mar-2019

Summary

Official Symbol	Tab2 provided by MGI
Official Full Name	TGF-beta activated kinase 1/MAP3K7 binding protein 2 provided by MGI
Primary source	MGI:MGI:1915902
See related	Ensembl:ENSMUSG0000015755
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	1110030N06Rik, A530078N03Rik, Map3k7ip2, mKIAA0733
Expression	Ubiquitous expression in limb E14.5 (RPKM 18.4), CNS E11.5 (RPKM 13.0) and 28 other tissues See more
Orthologs	human all

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Transcript information (Ensembl)



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The gene has 5 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tab2-204	ENSMUST00000146444.7	4380	<u>693aa</u>	Protein coding	CCDS23691	<u>Q99K90</u>	TSL:5 GENCODE basic APPRIS P1
Tab2-201	ENSMUST00000130322.1	2311	<u>385aa</u>	Protein coding	78	D3Z564	CDS 3' incomplete TSL:1
Tab2-205	ENSMUST00000147938.1	1980	<u>553aa</u>	Protein coding	10	D3Z216	CDS 3' incomplete TSL:5
Tab2-203	ENSMUST00000143848.1	703	No protein	Retained intron	20	100	TSL:2
Tab2-202	ENSMUST00000142007.1	618	No protein	Retained intron	5	(5)	TSL:2

The strategy is based on the design of *Tab2-204* transcript, The transcription is shown below

< Tab2-204 protein coding

Reverse strand -

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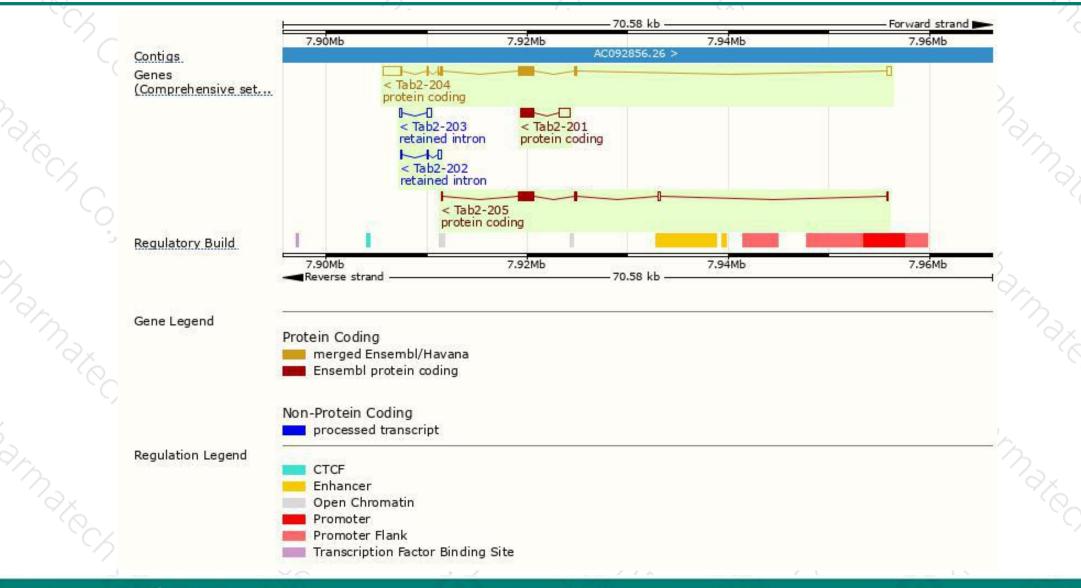
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50.58 kb

Genomic location distribution



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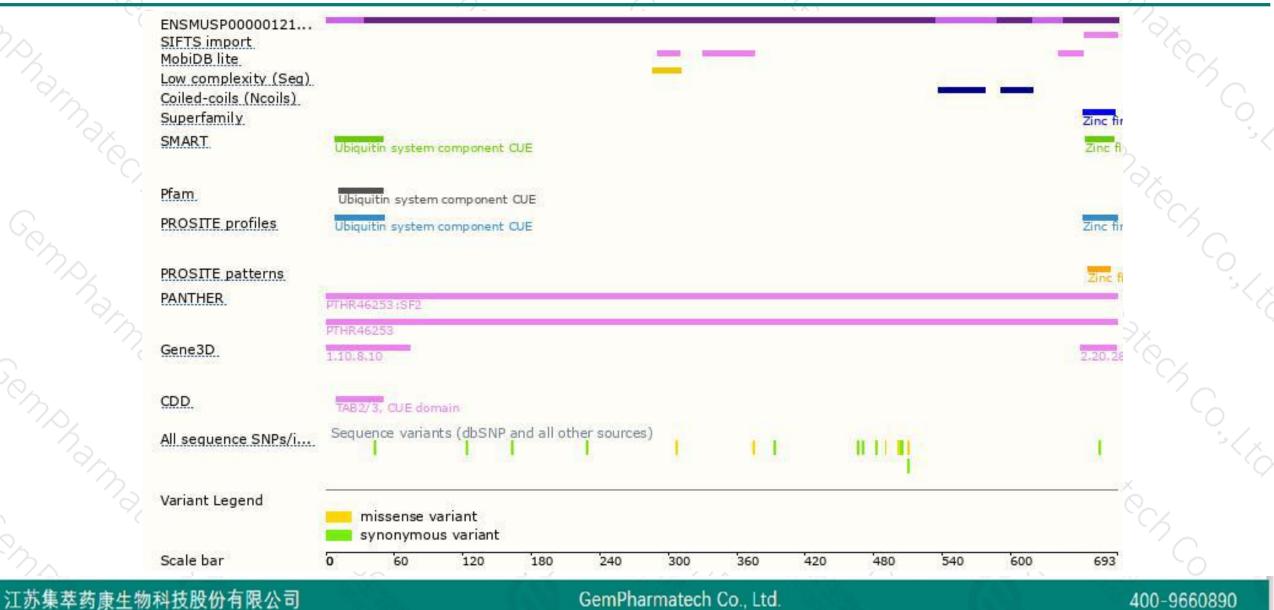


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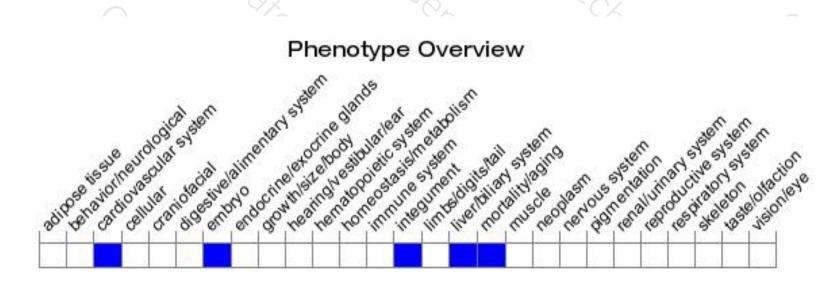
Protein domain





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, Embryos homozygous for a knock-out allele are viable up to E9.5. Embryos homozygous for a different knock-out allele are normal and viable up to E11.5 but become pale and anemic, exhibit liver hem and increased apoptosis of hepatoblasts, and die by E12.5.

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



