

Thumpd3 Cas9-KO Strategy

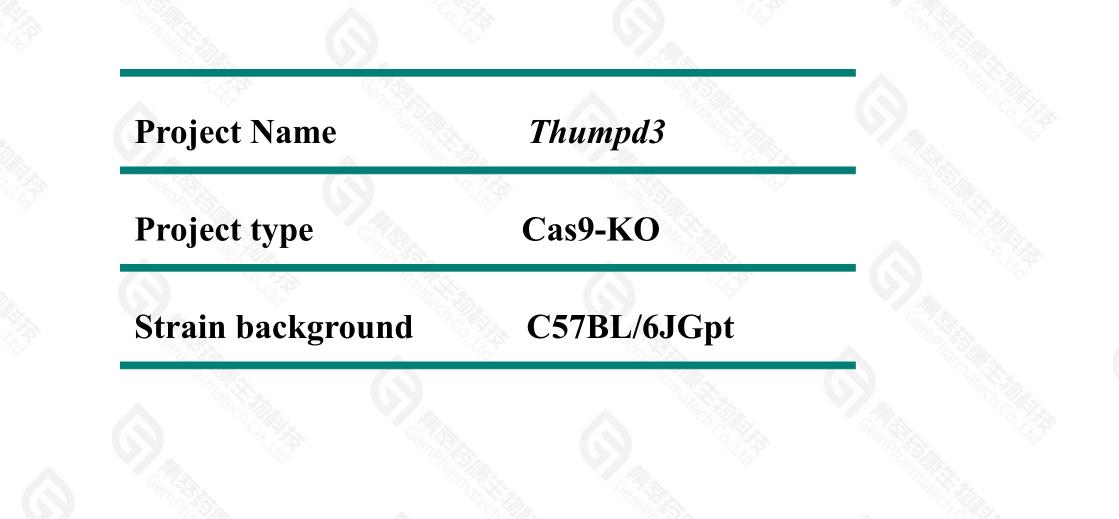
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Reviewer: Miaomiao Cui

Design Date: 2021-6-11

Project Overview





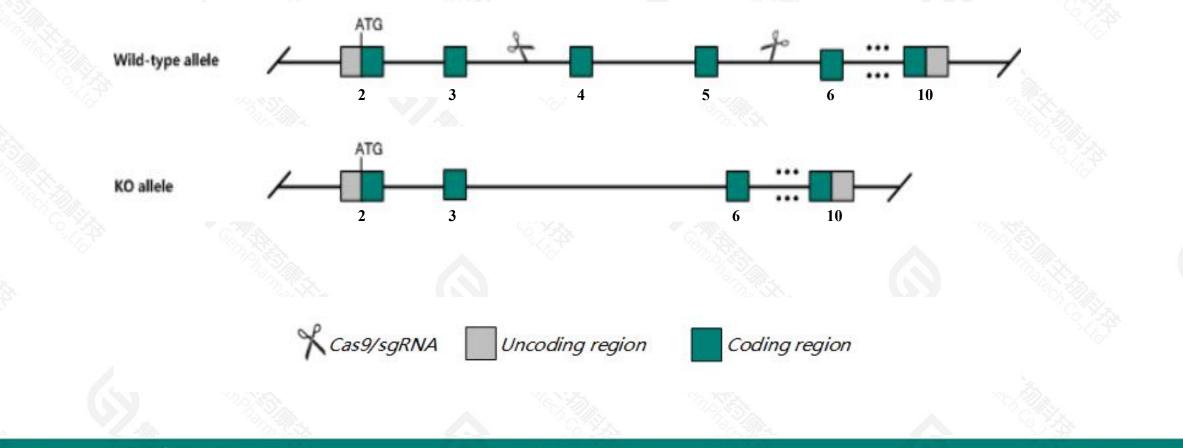
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Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Thumpd3* gene. The schematic diagram is as follows:



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> The *Thumpd3* gene has 10 transcripts. According to the structure of *Thumpd3* gene, exon4-exon5 of *Thumpd3*-201(ENSMUST00000032398.15) transcript is recommended as the knockout region. The region contains 608bp coding sequence. Knock out the region will result in disruption of protein function.

> In this project we use CRISPR/Cas9 technology to modify *Thumpd3* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.



- ➤ Transcript *Thumpd3*-204&210 may not be affected.
- > *Gm22591* may be deleted.

The *Thumpd3* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



☆ ?

Thumpd3 THUMP domain containing 3 [Mus musculus (house mouse)]

Gene ID: 14911, updated on 13-Mar-2020

- Summary

Official Symbol	Thumpd3 provided by MGI
Official Full Name	THUMP domain containing 3 provided by MGI
Primary source	MGI:MGI:1277973
See related	Ensembl:ENSMUSG00000030264
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; Mus; Mus
Also known as	AW556087, Gt(ROSA)26asSor, Gtrosa26as
Expression	Ubiquitous expression in testis adult (RPKM 10.3), placenta adult (RPKM 8.3) and 25 other tissues See more
Orthologs	human all

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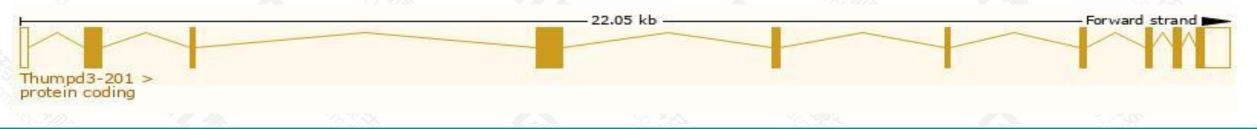
Transcript information (Ensembl)



The gene has 10 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Thumpd3-201	ENSMUST0000032398.14	2163	<u>505aa</u>	Protein coding	CCDS20410	P97770	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 P1
Thumpd3-208	ENSMUST00000155378.1	855	<u>221aa</u>	Protein coding	-	D3YYB2	CDS 3' incomplete TSL:2
Thumpd3-210	ENSMUST00000204866.2	727	<u>86aa</u>	Protein coding	-	A0A0N4SUS5	CDS 3' incomplete TSL:3
Thumpd3-204	ENSMUST00000138278.1	371	<u>77aa</u>	Protein coding	-	A0A0N4SW81	CDS 3' incomplete TSL:3
Thumpd3-202	ENSMUST00000134657.2	649	No protein	Processed transcript		-	TSL:2
Thumpd3-209	ENSMUST00000204786.1	3072	No protein	Retained intron	-	-	TSL:NA
Thumpd3-203	ENSMUST00000137396.7	1764	No protein	Retained intron	-	-	TSL:1
Thumpd3-207	ENSMUST00000153310.1	692	No protein	Retained intron	2	2	TSL:2
Thumpd3-206	ENSMUST00000152795.1	638	No protein	Retained intron			TSL:2
Thumpd3-205	ENSMUST00000145054.7	558	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Thumpd3-201* transcript, the transcription is shown below:

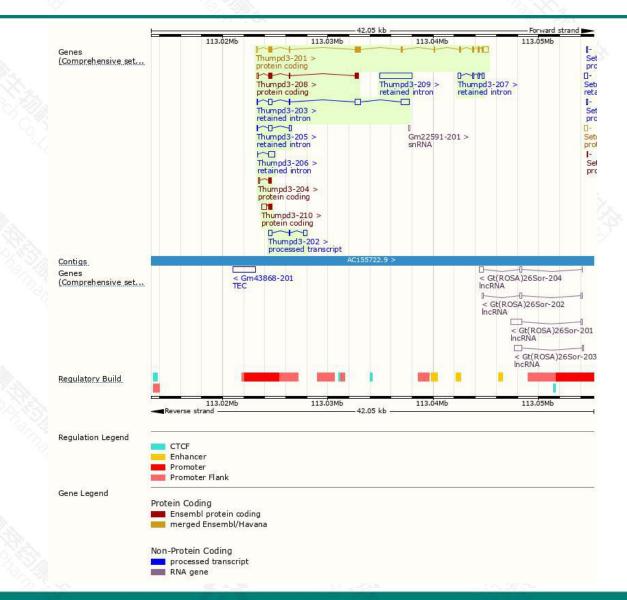


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Genomic location distribution



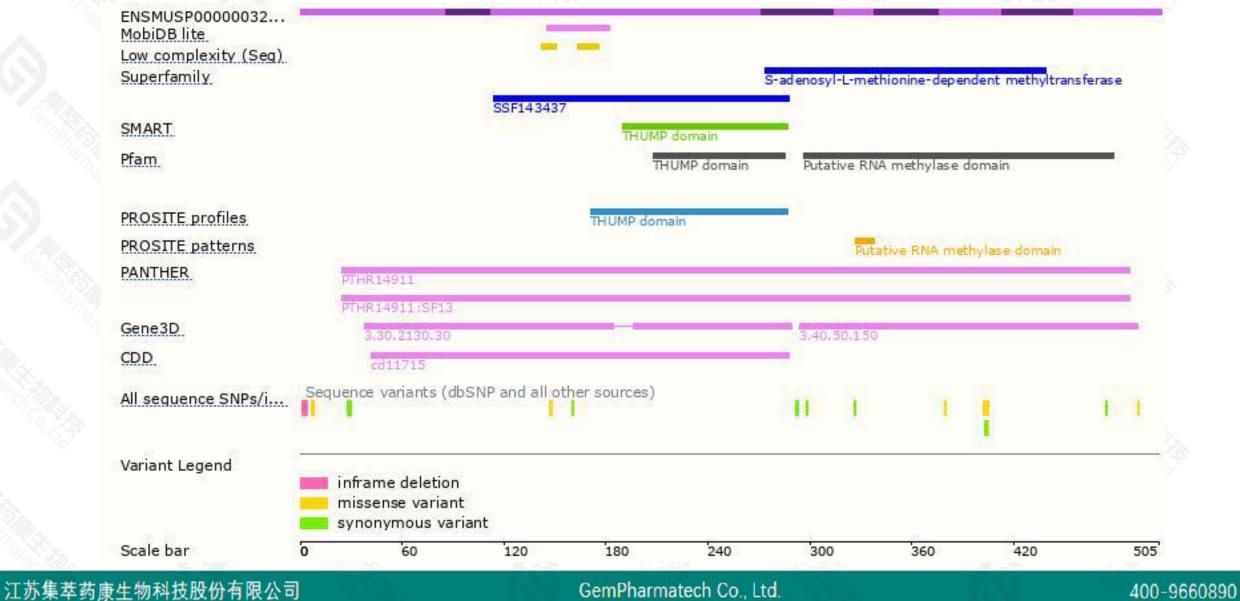


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Protein domain







If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



