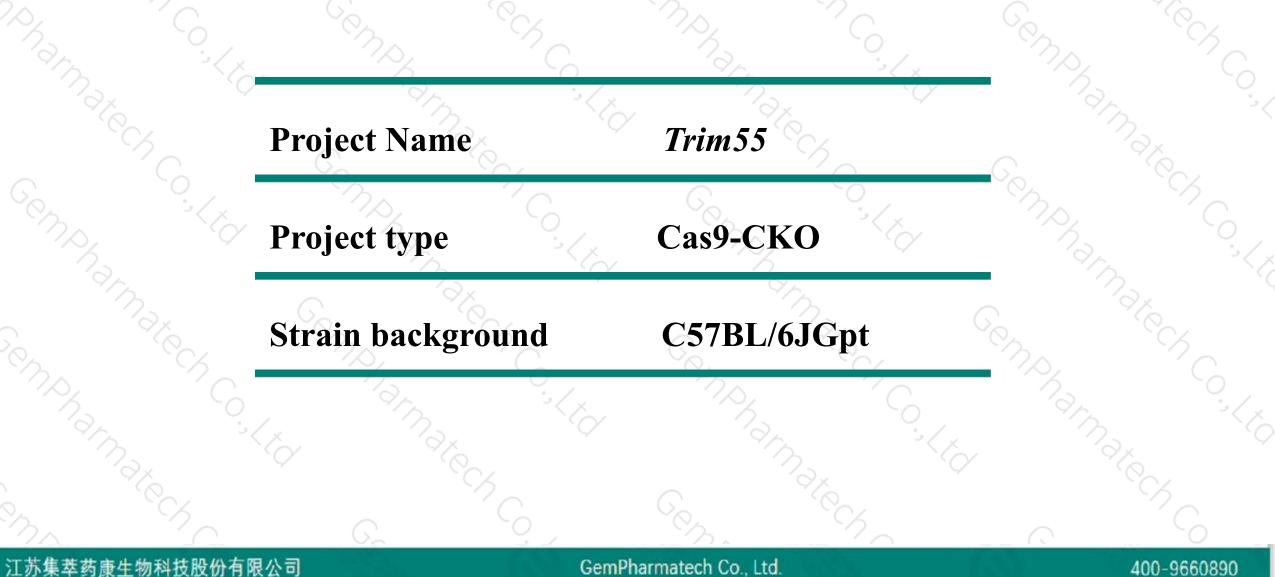


Trim55 Cas9-CKO Strategy

Designer: Reviewer: Design Date: Baocheng Zhuang Yang Zeng 2018-6-19

Project Overview





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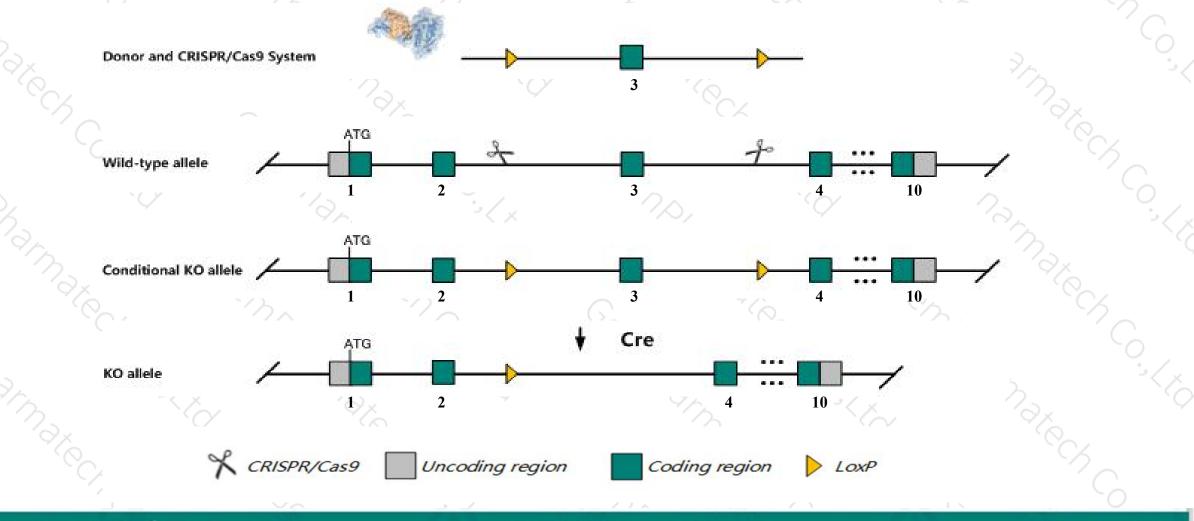
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Conditional Knockout strategy



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This model will use CRISPR/Cas9 technology to edit the *Trim55* gene. The schematic diagram is as follows:



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The Trim55 gene has 2 transcripts. According to the structure of Trim55 gene, exon3 of Trim55-201 (ENSMUST00000029139.8) transcript is recommended as the knockout region. The region contains 166bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Trim55* 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 null allele exhibit increased heart and muscle

to body weight ratios and cardiac hypertrophy.

- ➤ Transcript *Trim55-202* may not be affected.
- The Trim55 gene is located on the Chr3. 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)



\$?

Trim55 tripartite motif-containing 55 [Mus musculus (house mouse)]

Gene ID: 381485	, updated on	12-Aug-2019
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Summary

Official Symbol	Trim55 provided by MGI									
Official Full Name	tripartite motif-containing 55 provided by MGI									
Primary source	MGI:MGI:3036269									
See related	Ensembl:ENSMUSG0000060913									
Gene type	protein coding									
RefSeq status	VALIDATED									
Organism	Mus musculus									
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;									
	Myomorpha; Muroidea; Murinae; Mus; Mus									
Also known as	Murf2; Rnf29; D830041C10Rik									
Expression	Biased expression in heart adult (RPKM 18.9), limb E14.5 (RPKM 5.3) and 2 other tissues See more									
Orthologs	human all									
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Transcript information (Ensembl)



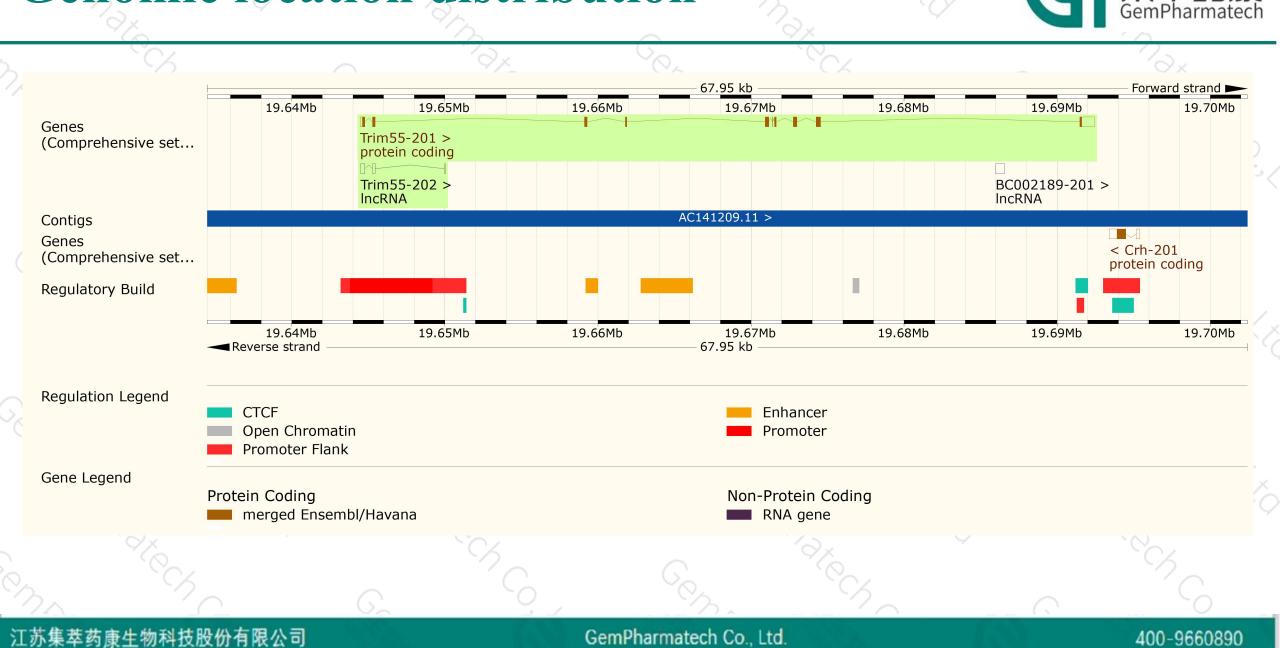
The gene has 2 transcripts, all transcripts are shown below:

Name 🔺	Transcript ID	bp 🌲	Protein 🝦	Translation ID	Biotype 🍦	CCDS 🔶	UniProt 🝦	Flags 🔶	
Trim55-201	ENSMUST0000029139.8	2595	<u>545aa</u>	ENSMUSP0000029139.7	Protein coding	<u>CCDS38399</u> &	<u>G3X8Y1</u>	TSL:2 GENCODE basic APPRIS P1	
Trim55-202	ENSMUST00000195744.1	506	No protein	-	IncRNA	-	×	TSL:3	

The strategy is based on the design of *Trim55-201* transcript, The transcription is shown below

TimEE 201 >	47.95 kb	Forward strand
Trim 55-201 > protein coding		
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		°°A C

Genomic location distribution

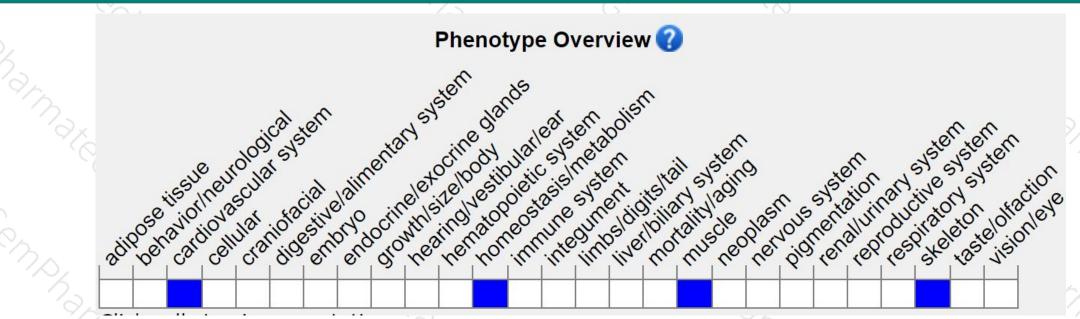


Protein domain



	5									
17	ENSMUSP00000029 MobiDB lite Low complexity (Seg) Superfamily	SSF57850	SSF57845		-			-	=	
	SMART	Zinc finger, RING-type	B-box-type zinc finger							
	Pfam	RING-type zinc-finger, LisH								
	PROSITE profiles	Zinc finger, RING-type	B-box-type zinc finger B-box-type zinc finger	COS	domain					
	PROSITE patterns PANTHER	Zinc finger, RING-type	, conserved site						<	
	PANTIER	PTHR24099								
	Gene3D	PTHR24099:SF17 Zinc finger, RING/FYVE/PHD-type	3.30.40.200	1.20.5.170						
	CDD	cd16760	B-box-type zinc finger							
	All sequence SNPs/i	Sequence variants (dbSNP and a	ll other sources)	н т. н	0				1	
1	Variant Legend	missense variant		-						
	Scale bar	0 60	120 180	240	300	360	420	480	545	
1	<u>`</u>			<u>`</u> ?>.		10			0	
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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 increased heart and muscle to body weight ratios and cardiac hypertrophy.

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



