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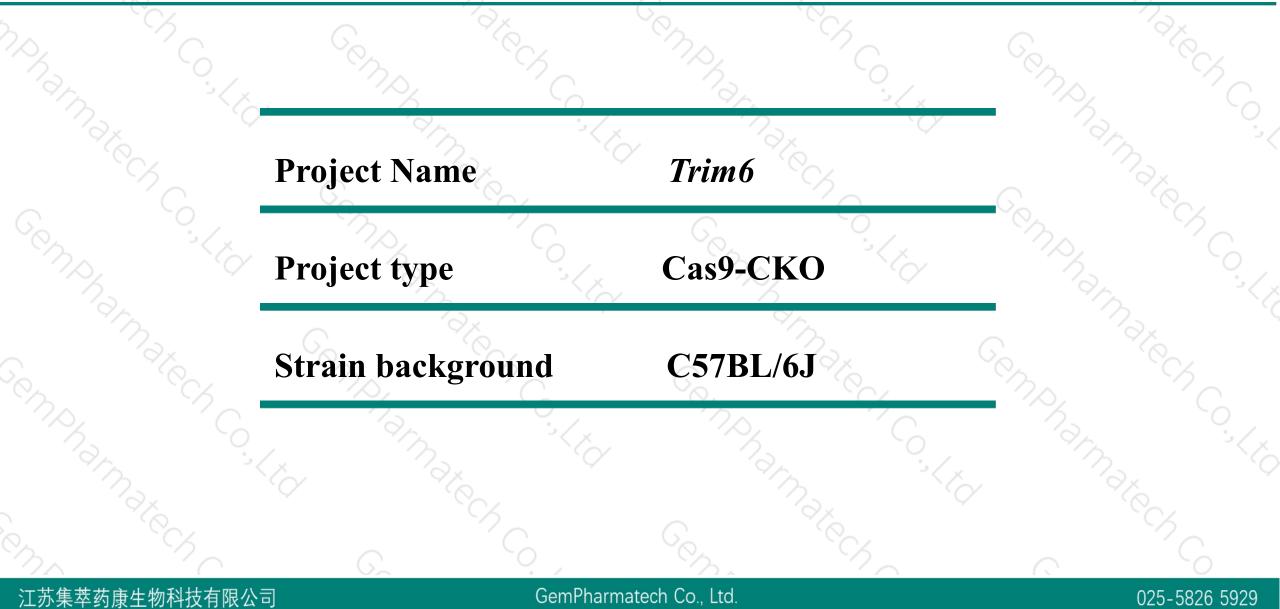
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Project Overview

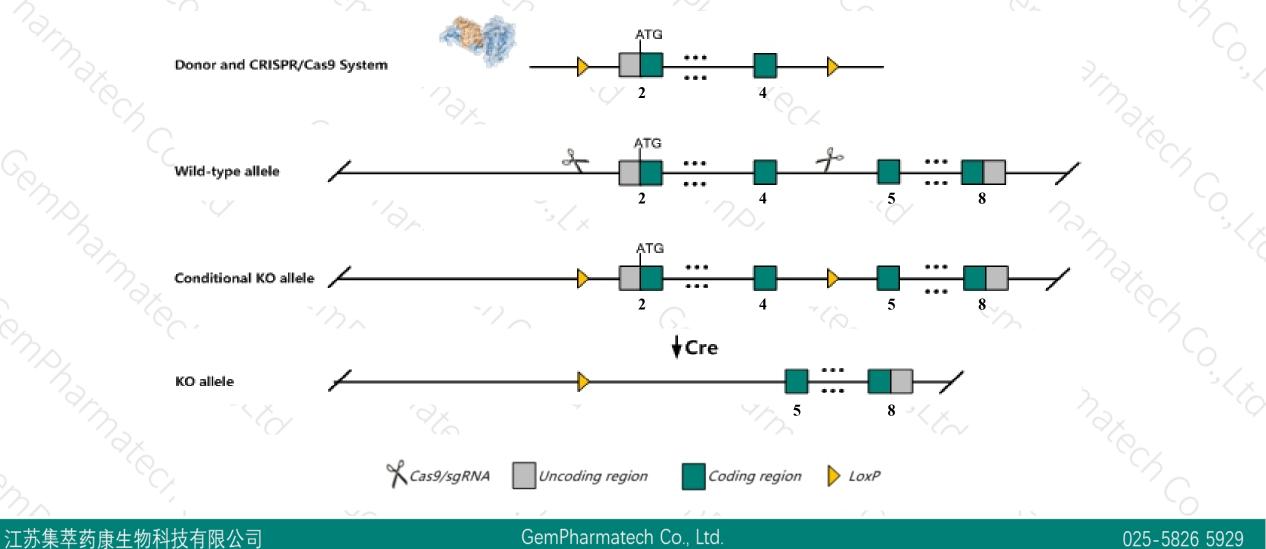




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Trim6* gene. The schematic diagram is as follows:





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The *Trim6* gene has 6 transcripts. According to the structure of *Trim6* gene, exon2-exon4 of *Trim6-201* (ENSMUST00000098180.9) 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 *Trim6* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA and Donor were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

The flox mice was 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.

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- The *Trim6* gene is located on the Chr7. 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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Trim6 tripartite motif-containing 6 [Mus musculus (house mouse)]

Gene ID: 94088, updated on 31-Jan-2019

Summary

Official Symbol	Trim6 provided by MGI
Official Full Name	tripartite motif-containing 6 provided by <u>MGI</u>
Primary source	MGI:MGI:2137352
See related	Ensembl:ENSMUSG00000072244
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	C430046K18Rik, D7Ertd684e
Expression	Biased expression in placenta adult (RPKM 14.8) and colon adult (RPKM 1.4)See more
Orthologs	human all

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Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

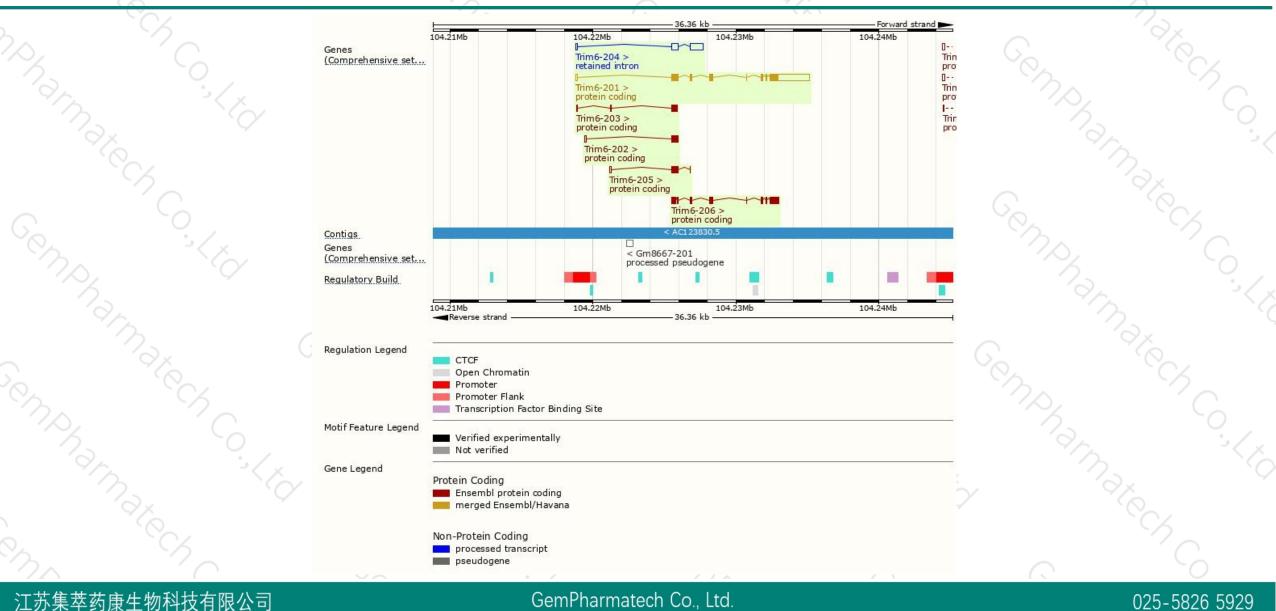
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
ENSMUST0000098180.9	3827	<u>488aa</u>	Protein coding	CCDS40065	Q8BGE7	TSL:1 GENCODE basic APPRIS P1	
ENSMUST00000214578.1	1506	<u>462aa</u>	Protein coding	-	A0A1L1SS93	TSL:5 GENCODE basic	
ENSMUST00000153324.1	622	<u>148aa</u>	Protein coding	-	D3Z7U8	CDS 3' incomplete TSL:3	
ENSMUST00000128493.1	536	<u>125aa</u>	Protein coding	-	<u>D3YY77</u>	CDS 3' incomplete TSL:3	
ENSMUST00000144455.1	520	<u>113aa</u>	Protein coding	-	<u>D3Z676</u>	CDS 3' incomplete TSL:3	
ENSMUST00000144538.1	1490	No protein	Retained intron	-	-	TSL:1	
	ENSMUST0000098180.9 ENSMUST00000214578.1 ENSMUST00000153324.1 ENSMUST00000128493.1 ENSMUST00000144455.1	ENSMUST0000098180.9 3827 ENSMUST00000214578.1 1506 ENSMUST00000153324.1 622 ENSMUST00000128493.1 536 ENSMUST00000144455.1 520	ENSMUST0000098180.9 3827 488aa ENSMUST00000214578.1 1506 462aa ENSMUST0000153324.1 622 148aa ENSMUST0000128493.1 536 125aa ENSMUST00000144455.1 520 113aa	ENSMUST0000098180.93827488aaProtein codingENSMUST00000214578.11506462aaProtein codingENSMUST00000153324.1622148aaProtein codingENSMUST00000128493.1536125aaProtein codingENSMUST00000144455.1520113aaProtein coding	ENSMUST0000098180.93827488aaProtein codingCCDS40065ENSMUST00000214578.11506462aaProtein coding-ENSMUST0000153324.1622148aaProtein coding-ENSMUST00000128493.1536125aaProtein coding-ENSMUST00000144455.1520113aaProtein coding-	ENSMUST0000098180.93827488aaProtein codingCCDS40065Q8BGE7ENSMUST0000214578.11506462aaProtein coding-A0A1L1SS93ENSMUST0000153324.1622148aaProtein coding-D3Z7U8ENSMUST0000128493.1536125aaProtein coding-D3YY77ENSMUST0000144455.1520113aaProtein coding-D3Z676	

The strategy is based on the design of *Trim6-201* transcript, The transcription is shown below

Trim6-201 > protein coding			16.	36 kb		For	rward strand
70	('	20	· /	(D.)	10	63	$\overline{\mathbf{U}}$
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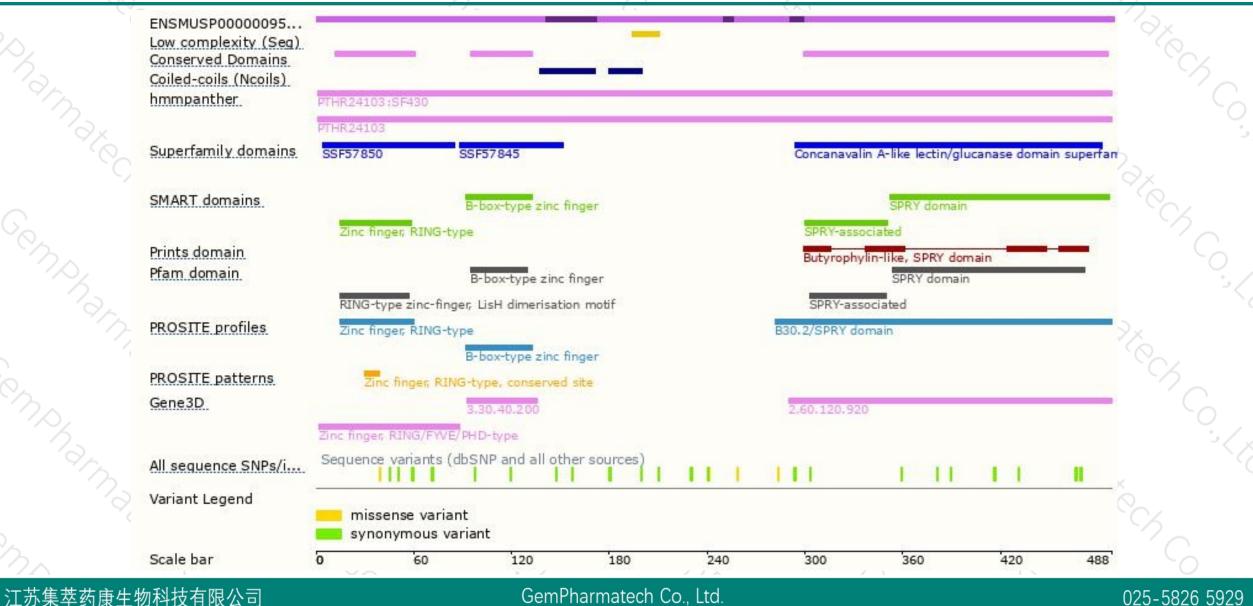
Genomic location distribution





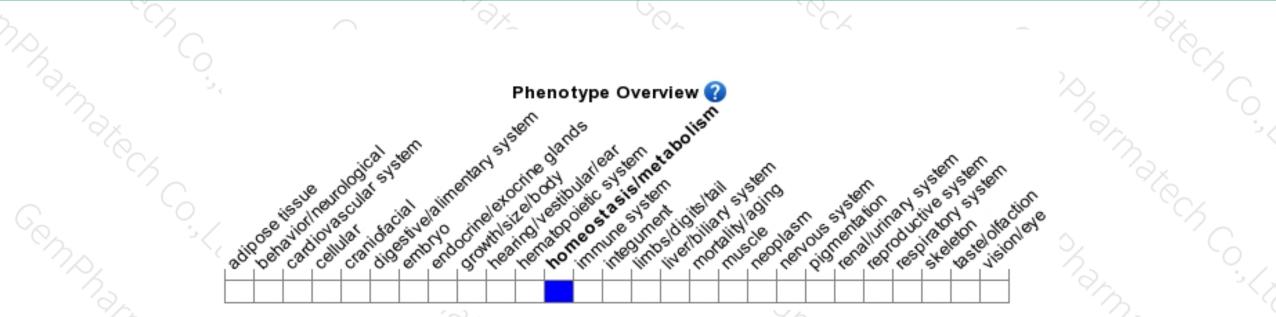
Protein domain





Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).



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



