

ND AMARCH CHILL Ripk1 Cas9-CKO Strategy Romphamater Control

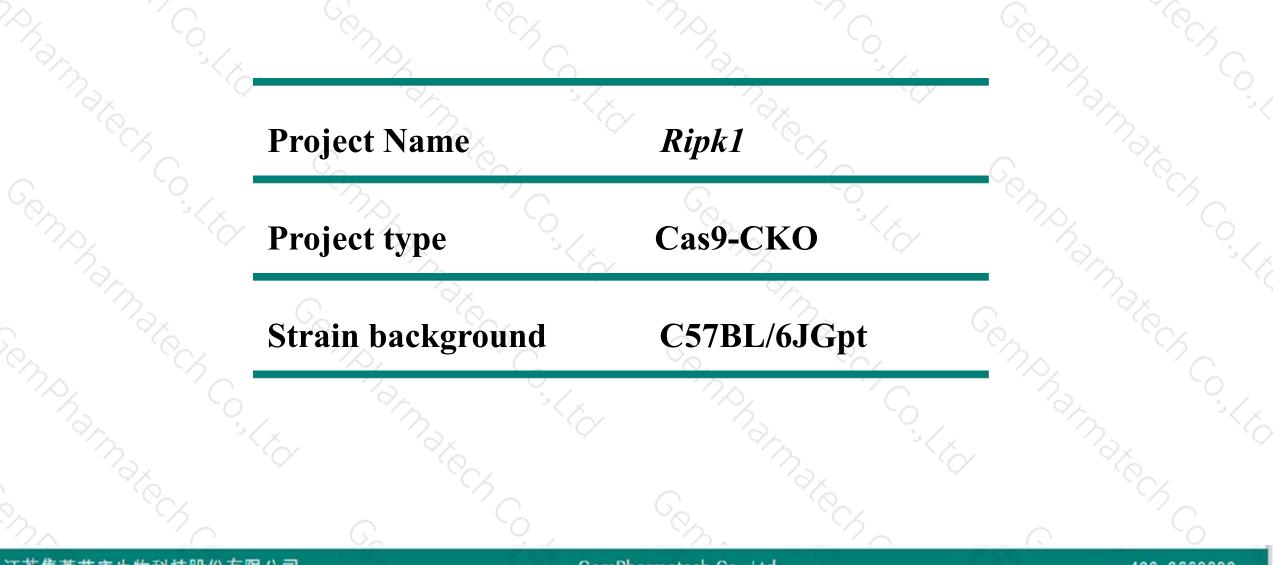
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Project Overview





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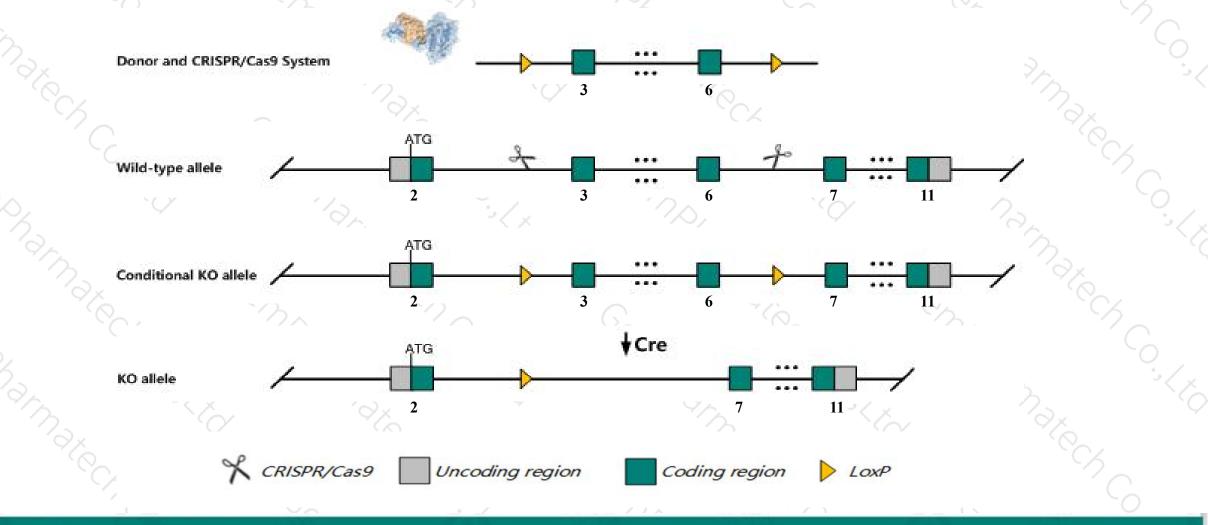
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Conditional Knockout strategy



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This model will use CRISPR/Cas9 technology to edit the *Ripk1* gene. The schematic diagram is as follows:



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The *Ripk1* gene has 5 transcripts. According to the structure of *Ripk1* gene, exon3-exon6 of *Ripk1-203* (ENSMUST00000167374.8) transcript is recommended as the knockout region. The region contains 677bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Ripk1* 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 disruptions in this gene die within 1 and 3 days of birth displaying extensive apoptosis in both lymphoid and adipose tissue.
- The *Ripk1* gene is located on the Chr13. 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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Ripk1 receptor (TNFRSF)-interacting serine-threonine kinase 1 [Mus musculus (house mouse)]

Gene ID: 19766, updated on 9-Apr-2019

Summary

Official Symbol	Ripk1 provided by MGI
Official Full Name	receptor (TNFRSF)-interacting serine-threonine kinase 1 provided by MGI
Primary source	MGI:MGI:108212
See related	Ensembl:ENSMUSG0000021408
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	D330015H01Rik, RIP, RIP-1, Rinp, Rip1
Expression	Ubiquitous expression in ovary adult (RPKM 4.7), lung adult (RPKM 4.4) and 28 other tissues See more
Orthologs	human all

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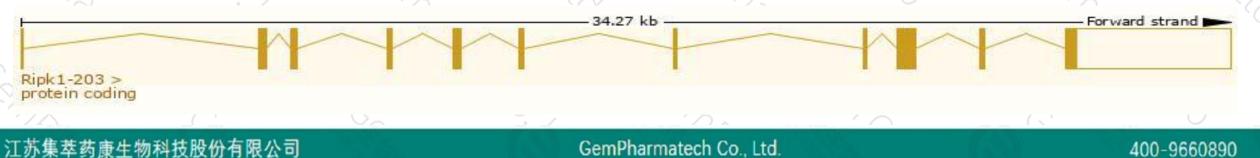
Transcript information (Ensembl)



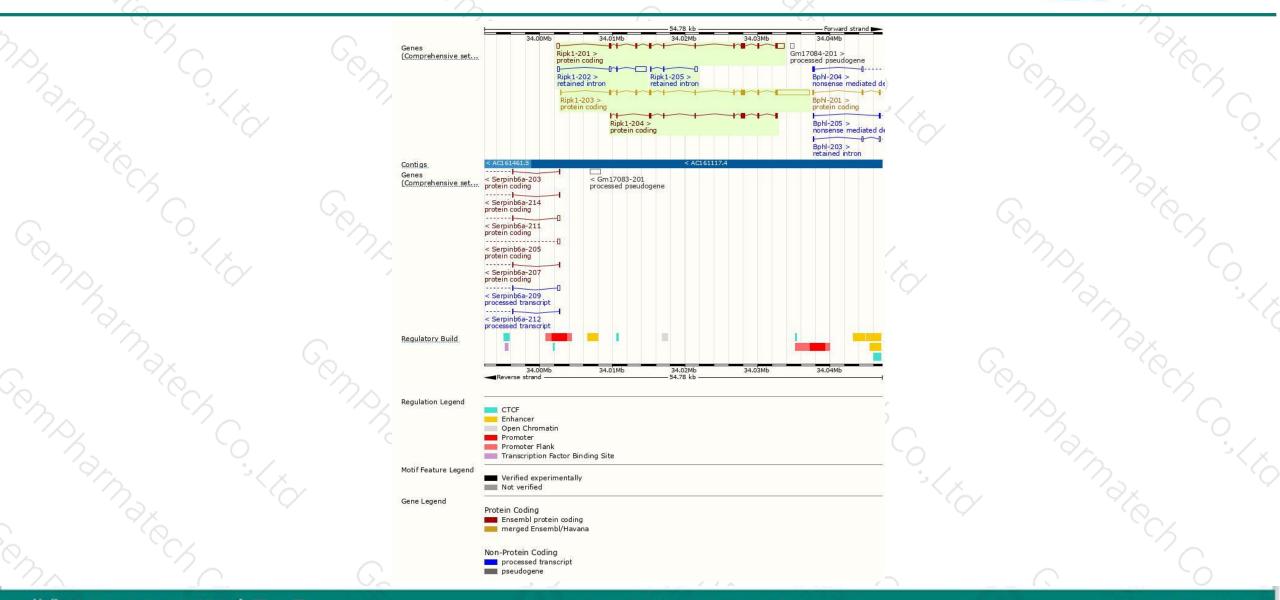
The gene has 5 transcripts, all transcripts are shown below:

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Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
Ripk1-203	ENSMUST00000167374.8	6472	<u>656aa</u>	Protein coding	CCDS26443	<u>Q60855</u>	TSL:1 GENCODE basic APPRIS P1	
Ripk1-201	ENSMUST00000021844.14	3266	<u>656aa</u>	Protein coding	CCDS26443	<u>Q60855</u>	TSL:1 GENCODE basic APPRIS P1	
Ripk1-204	ENSMUST00000171137.1	1792	<u>597aa</u>	Protein coding	34	<u>F7D1J2</u>	CDS 5' incomplete TSL:5	
Ripk1-202	ENSMUST00000163418.1	2121	No protein	Retained intron	12	24	TSL:1	
Ripk1-205	ENSMUST00000222176.1	570	No protein	Retained intron	67		TSL:3	

The strategy is based on the design of *Ripk1-203* transcript, The transcription is shown below



Genomic location distribution



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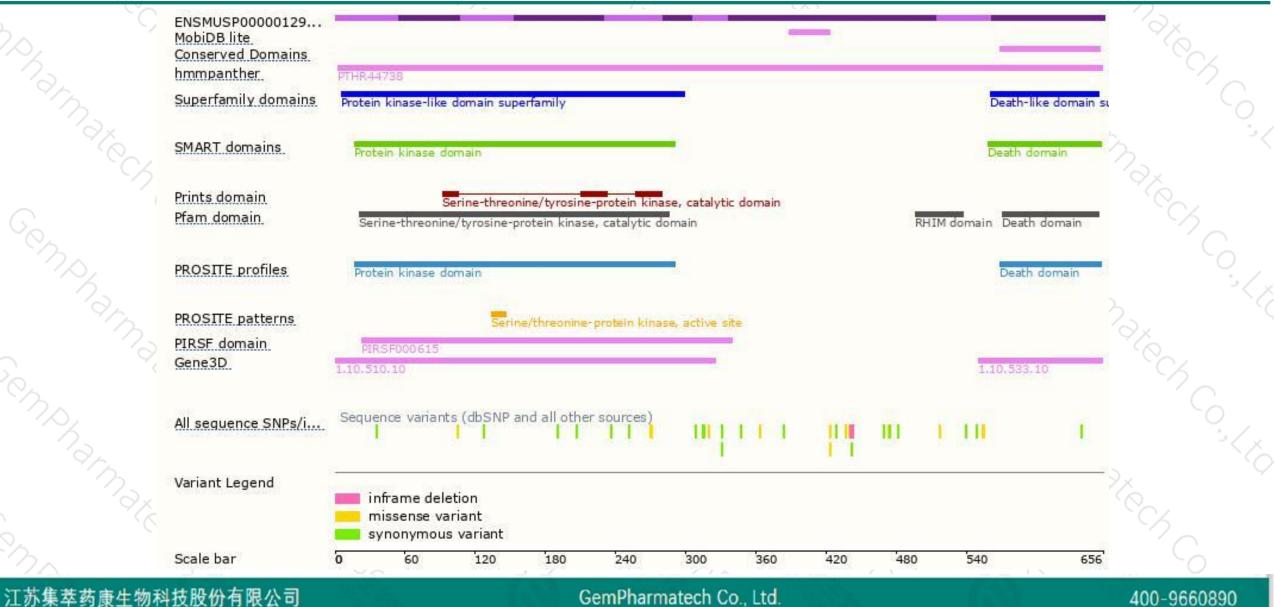
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Protein domain





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, Mice homozygous for disruptions in this gene die within 1 and 3 days of birth displaying extensive apoptosis in both lymphoid and adipose tissue.



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



