

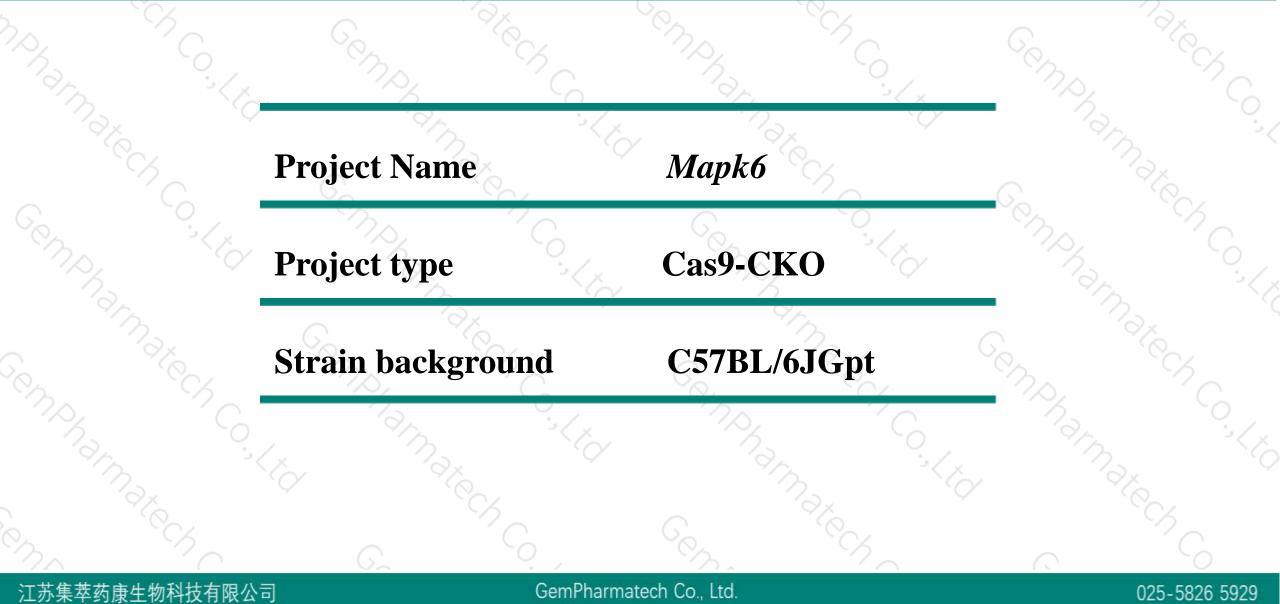
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Mapk6 Cas9-CKO Strategy Enphamaten Co-14

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Project Overview

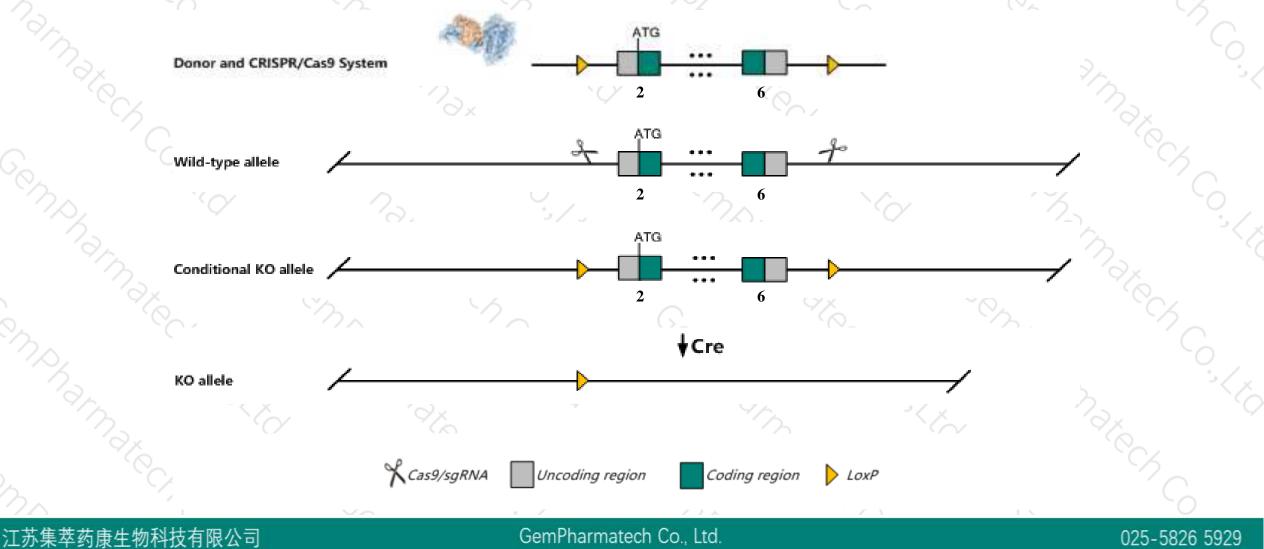




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Mapk6* gene. The schematic diagram is as follows:





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- The Mapk6 gene has 7 transcripts. According to the structure of Mapk6 gene, exon2-exon6 of Mapk6-202 (ENSMUST00000168937.7) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Mapk6* 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/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 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.



- According to the existing MGI data, homozygous null mice show limited fetal growth, reduced serum igf2 levels, pulmonary hypoplasia and early neonatal death. about 40% of newborns die of acute respiratory failure exhibiting delayed lung maturation, reduced sacculation, atelectasis, and impaired type ii pneumocyte differentiation.
- The Mapk6 gene is located on the Chr9. 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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Mapk6 mitogen-activated protein kinase 6 [Mus musculus (house mouse)]

Gene ID: 50772, updated on 13-Mar-2020

Summary

Official Symbol	Mapk6 provided by MGI
Official Full Name	mitogen-activated protein kinase 6 provided by MGI
Primary source	MGI:MGI:1354946
See related	Ensembl:ENSMUSG0000042688
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	2610021I23Rik, D130053K17Rik, Erk3, Mapk4, Mapk63, Prkm4, Prkm6
Expression	Broad expression in testis adult (RPKM 62.3), CNS E11.5 (RPKM 31.4) and 25 other tissuesSee more
Orthologs	human all

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Transcript information (Ensembl)



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The gene has 7 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mapk6-202	ENSMUST00000168937.7	4089	<u>720aa</u>	Protein coding	CCDS23342	<u>Q61532</u>	TSL:5 GENCODE basic APPRIS P1
Mapk6-201	ENSMUST0000049355.10	4072	<u>720aa</u>	Protein coding	CCDS23342	<u>Q61532</u>	TSL:1 GENCODE basic APPRIS P1
Mapk6-206	ENSMUST00000173460.1	875	No protein	Processed transcript	121	122	TSL:2
Mapk6-203	ENSMUST00000172665.1	471	No protein	Processed transcript	1970	2.55	TSL:3
Mapk6-205	ENSMUST00000173230.1	265	No protein	Processed transcript	2340	144	TSL:5
Mapk6-204	ENSMUST00000172946.1	3228	No protein	Retained intron	850		TSL:1
Mapk6-207	ENSMUST00000174034.1	615	No protein	Retained intron	(c)		TSL:2

The strategy is based on the design of Mapk6-202 transcript, the transcription is shown below:

< Mapk6-202 protein coding

Reverse strand

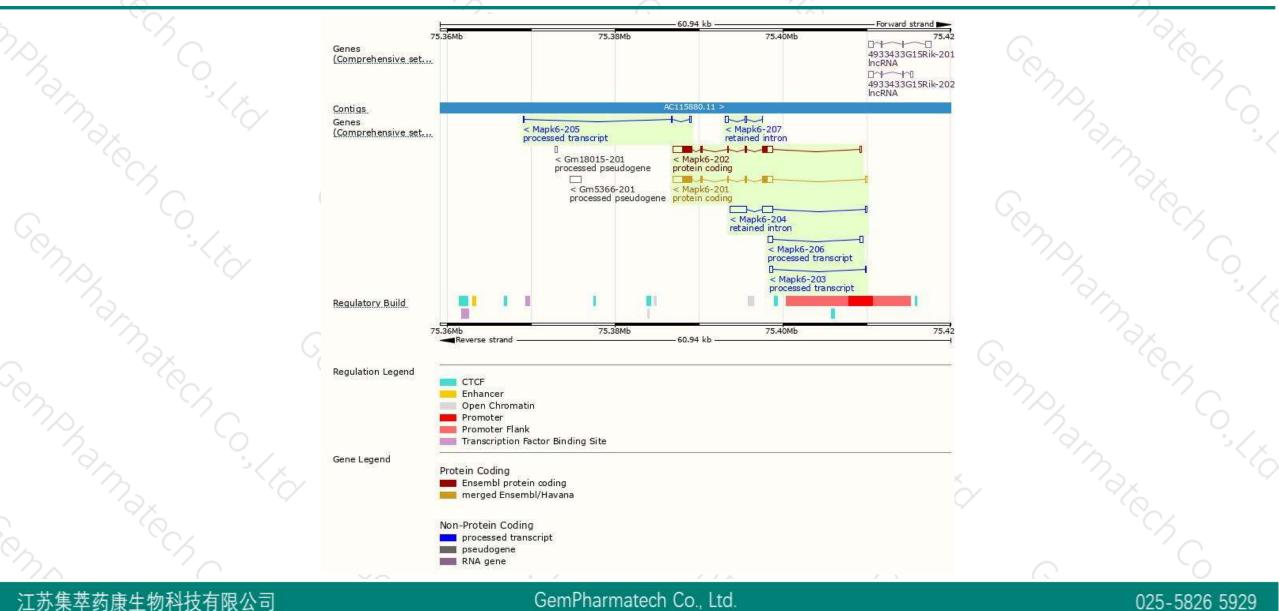
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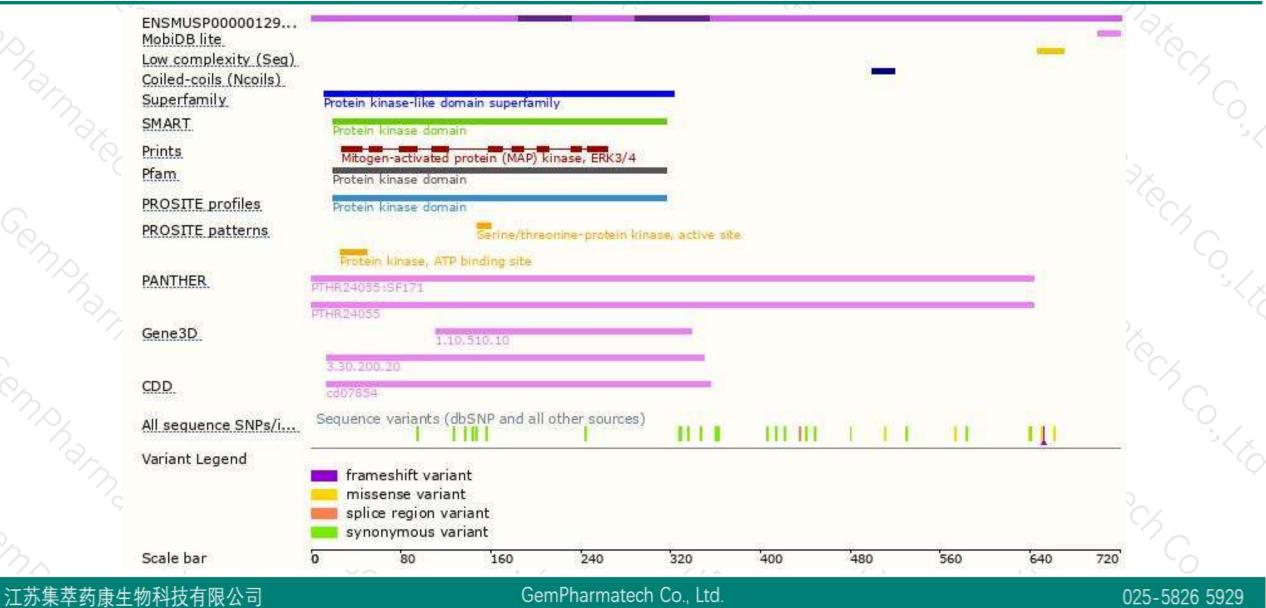
Genomic location distribution





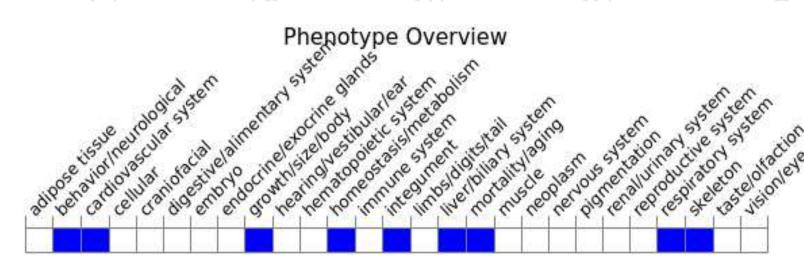
Protein domain





Mouse phenotype description(MGI)





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

According to the existing MGI data, homozygous null mice show limited fetal growth, reduced serum IGF2 levels, pulmonary hypoplasia and early neonatal death. About 40% of newborns die of acute respiratory failure exhibiting delayed lung maturation, reduced sacculation, atelectasis, and impaired type II pneumocyte differentiation.

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



