

# Gata4 Cas9-CKO Strategy

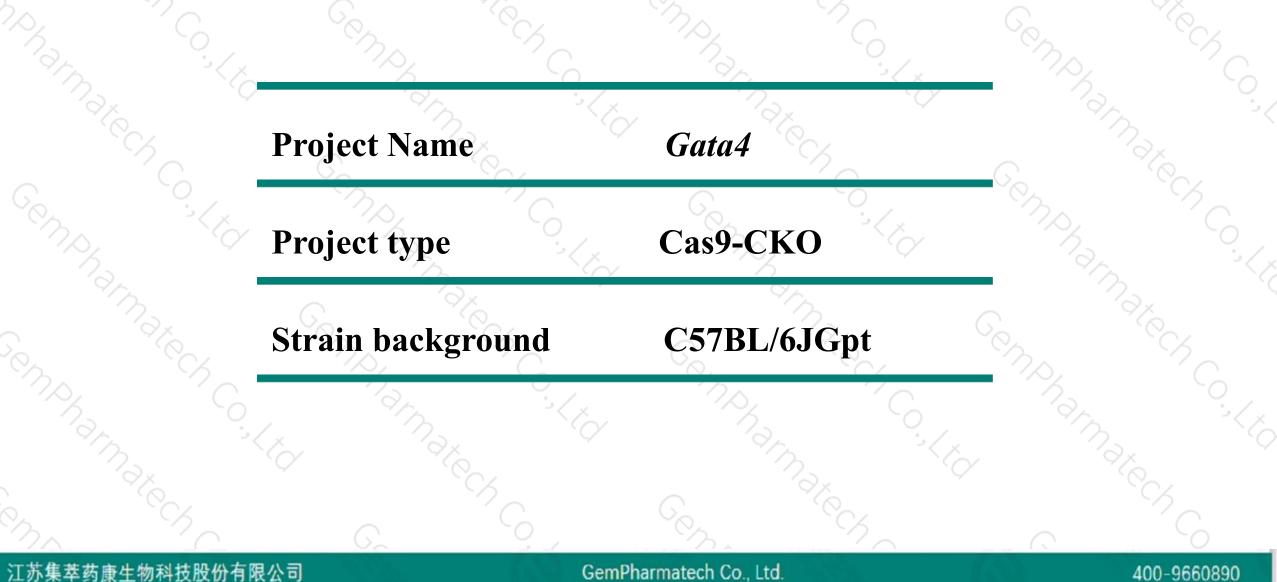
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Yupeng Yang 2019-7-23

# **Project Overview**





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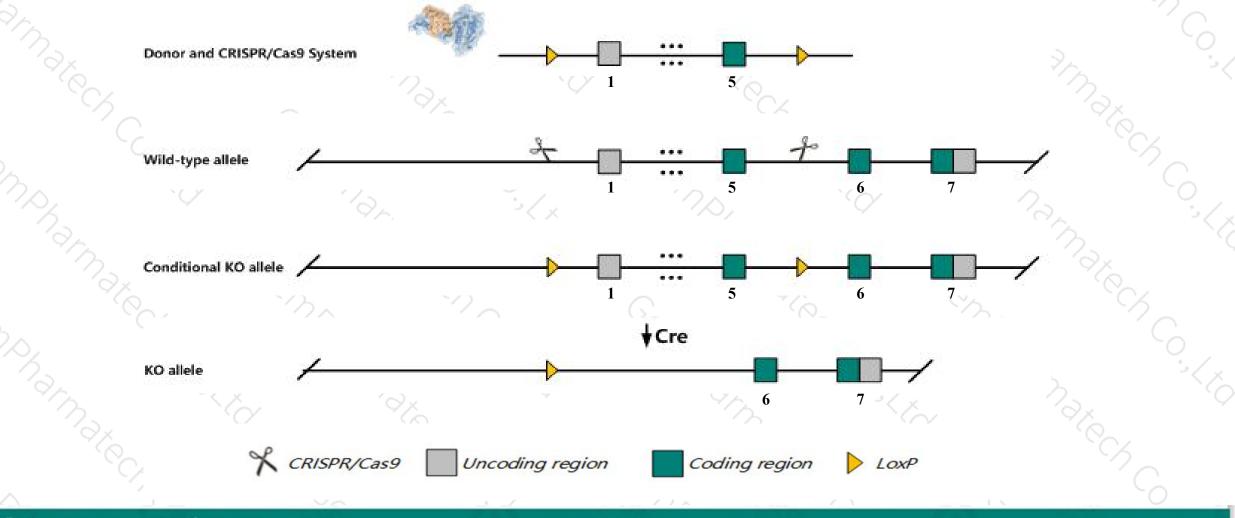
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# **Conditional Knockout strategy**



400-9660890

This model will use CRISPR/Cas9 technology to edit the *Gata4* gene. The schematic diagram is as follows:



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The Gata4 gene has 6 transcripts. According to the structure of Gata4 gene, exon1-exon5 of Gata4-201 (ENSMUST00000067417.9) transcript is recommended as the knockout region. The region contains 997bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Gata4* 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, Homozygotes for targeted null mutations exhibit defects in ventral morphogenesis, lack a primitive heart tube and foregut, develop partially outside the yolk sac, and die by midgestation.
- The Gata4 gene is located on the Chr14. 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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#### Gata4 GATA binding protein 4 [Mus musculus (house mouse)]

Gene ID: 14463, updated on 23-Mar-2019

#### Summary

Official SymbolGata4 provided by MGIOfficial Full NameGATA binding protein 4 provided by MGIPrimary soureMcl:MGI:95664See relateEnsembl:ENSMUSG0000021944Gene typeprotein codingprotein codingVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Rodentia; Myomorpha;<br/>Muroidea; Murinae; Mus; MusAlso knowansGata-4ExpressionBiased expression in ovary adult (RPKM 86.2), stomach adult (RPKM 63.9) and 5 other tissues<br/>Mum adult

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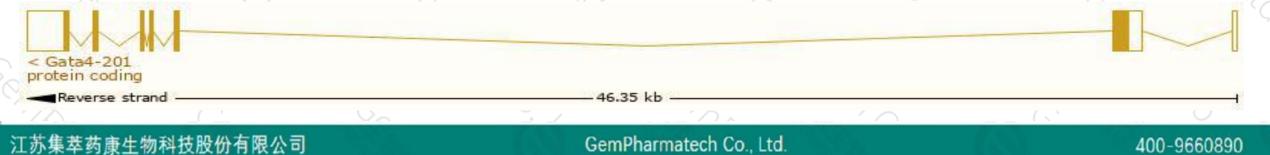
# **Transcript information (Ensembl)**



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

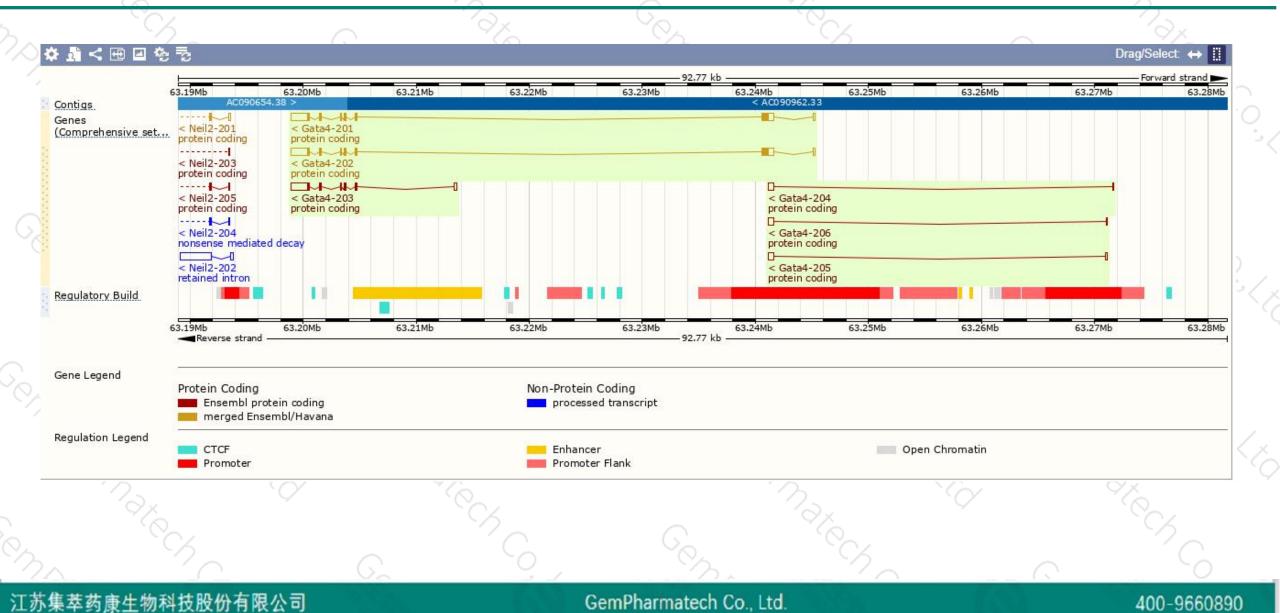
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Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gata4-201	ENSMUST0000067417.9	3400	<u>442aa</u>	Protein coding	CCDS79331	E9PXW9	TSL:1 GENCODE basic APPRIS ALT2
Gata4-202	ENSMUST00000118022.7	3374	<u>441aa</u>	Protein coding	CCDS49519	<u>Q08369</u>	TSL:1 GENCODE basic APPRIS P3
Gata4-203	ENSMUST00000121312.1	2407	<u>236aa</u>	Protein coding	-	Q3UYJ1	TSL:1 GENCODE basic
Gata4-205	ENSMUST00000137244.7	683	<u>11aa</u>	Protein coding	24	<u>A4K4Z6</u>	CDS 3' incomplete TSL:1
Gata4-204	ENSMUST00000132122.1	653	<u>15aa</u>	Protein coding	5	D3YTQ2	CDS 3' incomplete TSL:1
Gata4-206	ENSMUST00000156782.1	599	<u>11aa</u>	Protein coding		<u>A4K4Z6</u>	CDS 3' incomplete TSL:1

The strategy is based on the design of Gata4-201 transcript, The transcription is shown below



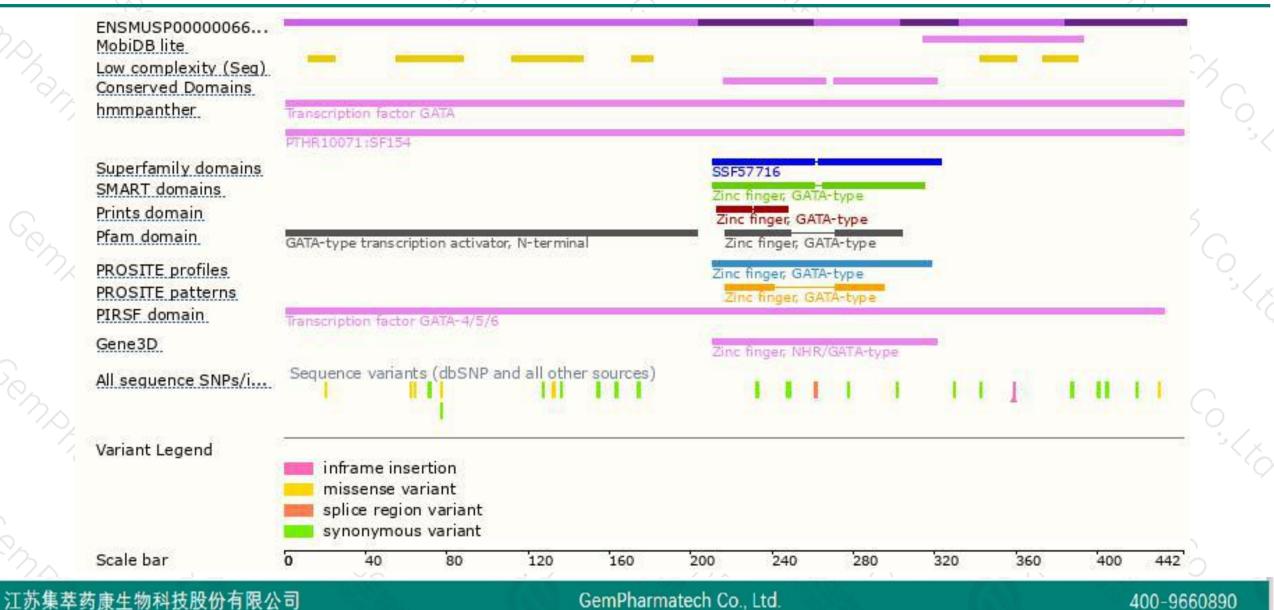
## **Genomic location distribution**





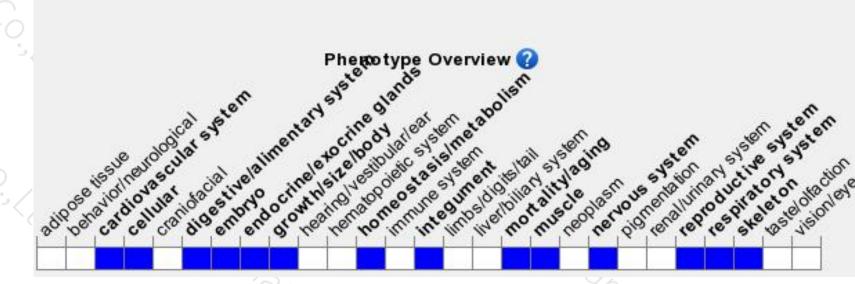
### **Protein domain**





# 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, Homozygotes for targeted null mutations exhibit defects in ventral morphogenesis, lack a primitive heart tube and foregut, develop partially outside the yolk sac, and die by midgestation.



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



