

Gata4 Cas9-CKO Strategy

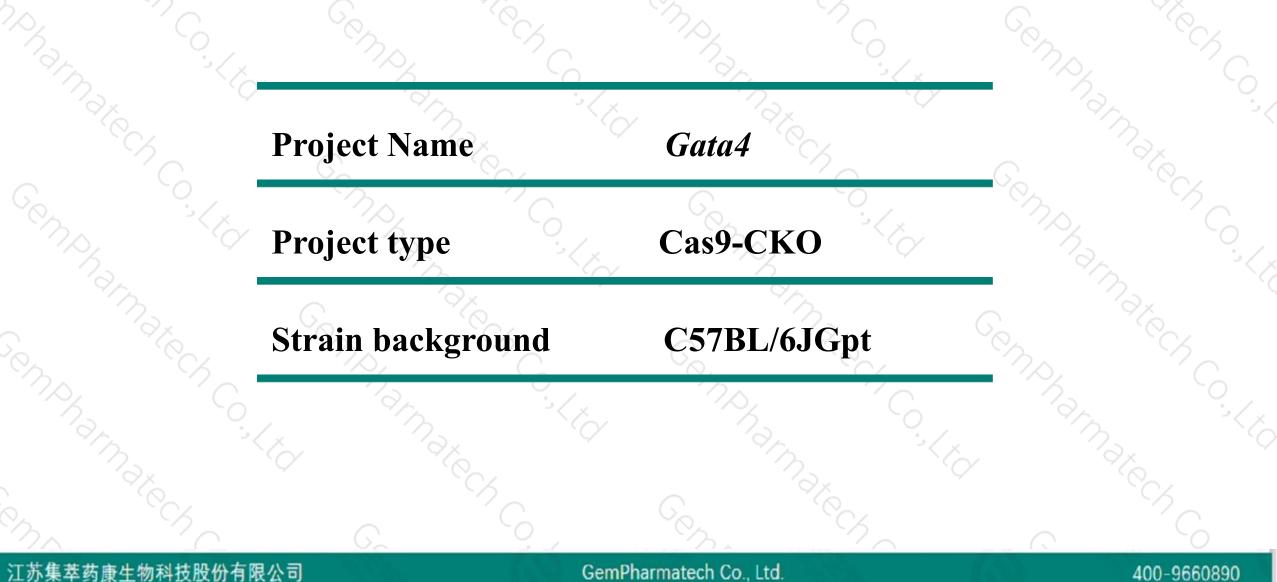
Designer: Design Date:

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Yupeng Yang 2019-7-23

Project Overview





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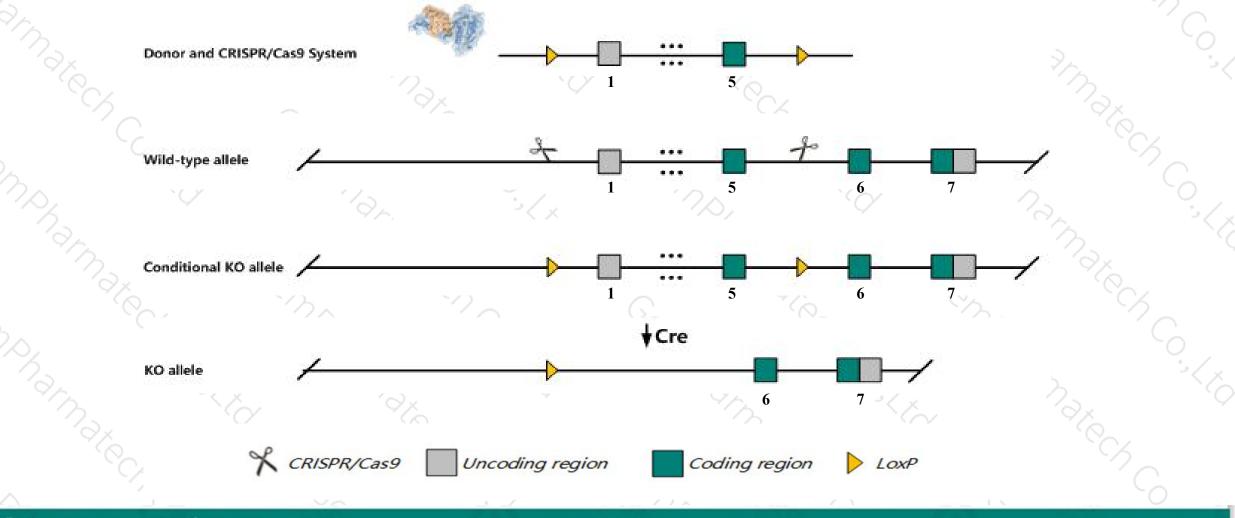
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Conditional Knockout strategy



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)



☆ ?

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;
Muroidea; Murinae; Mus; MusAlso knowansGata-4ExpressionBiased expression in ovary adult (RPKM 86.2), stomach adult (RPKM 63.9) and 5 other tissues
Mum adult

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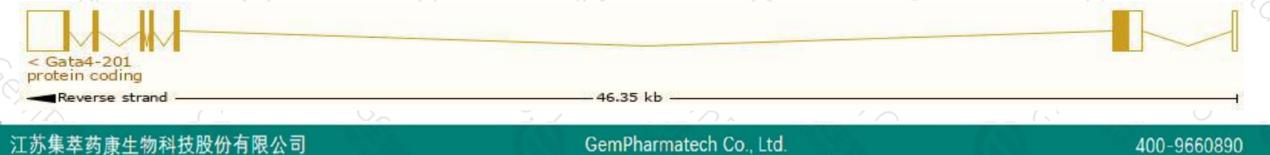
Transcript information (Ensembl)



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

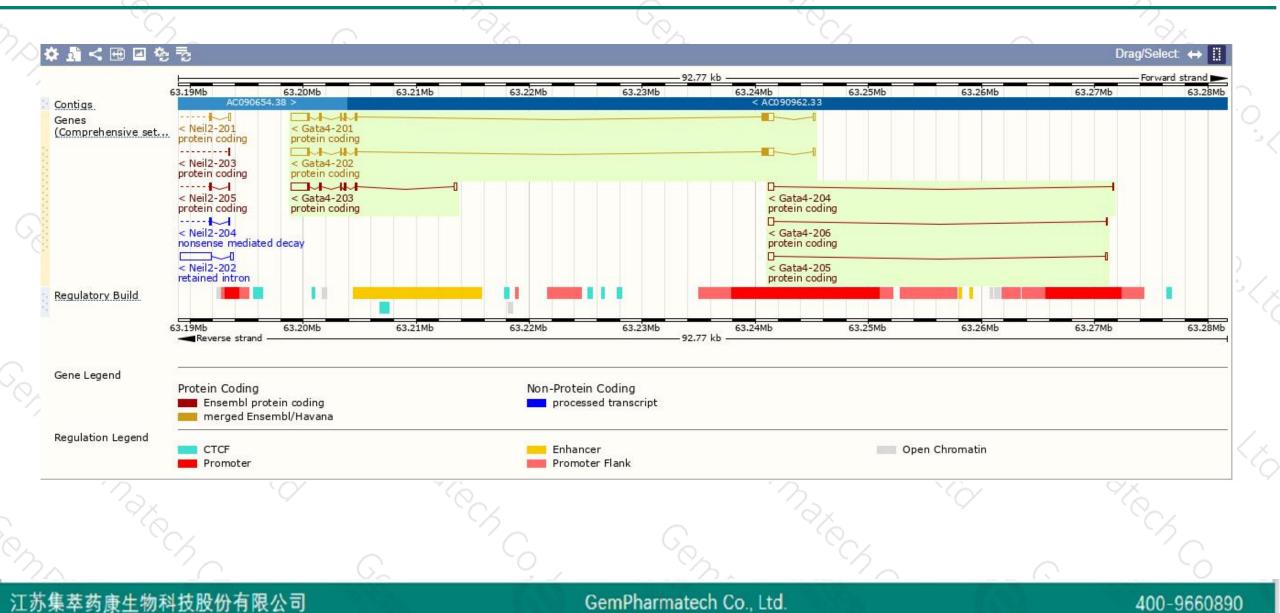
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|-----------|----------------------|------|--------------|---|-----------|---------------|---------------------------------|
| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
| Gata4-201 | ENSMUST0000067417.9 | 3400 | <u>442aa</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



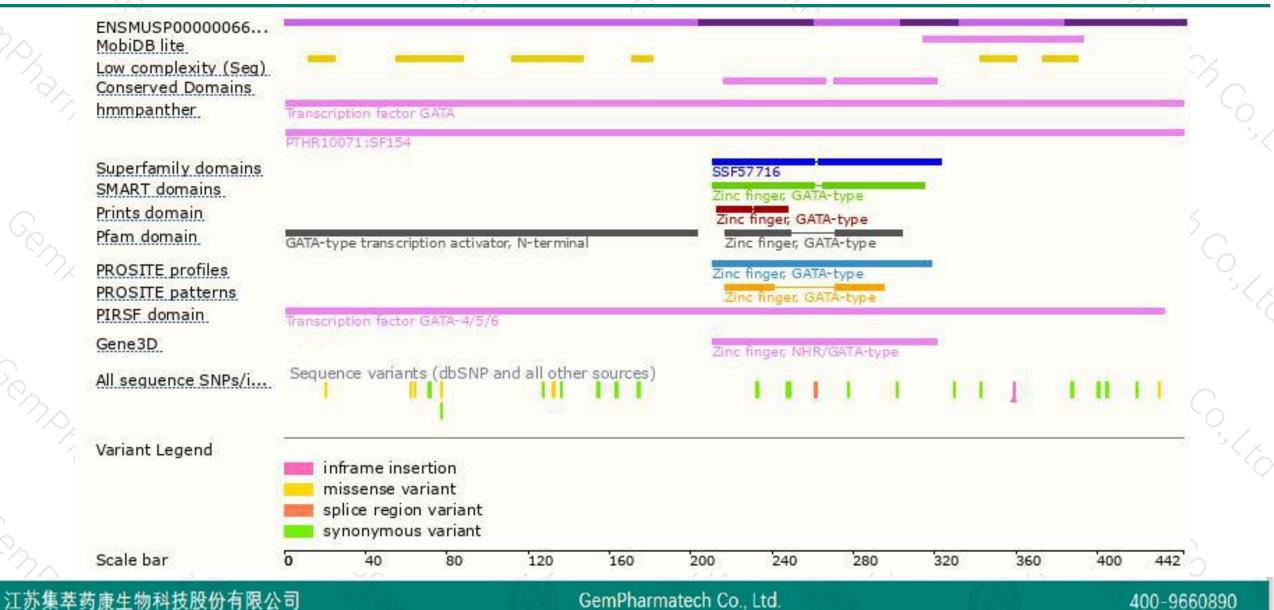
Genomic location distribution





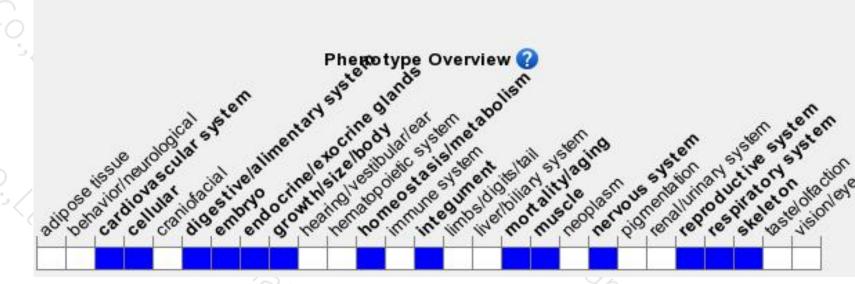
Protein domain





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



