

Gnb4 Cas9-CKO Strategy

Designer: Reviewer:

Design Date:

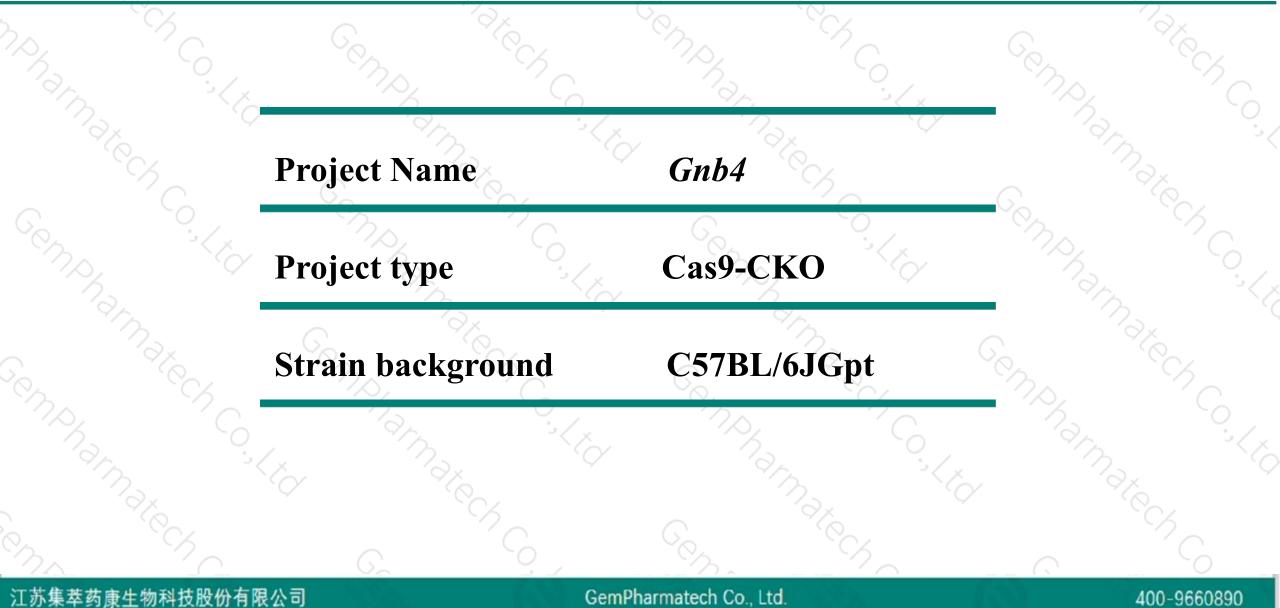
Ruirui Zhang

Huimin Su

2020-2-19

Project Overview



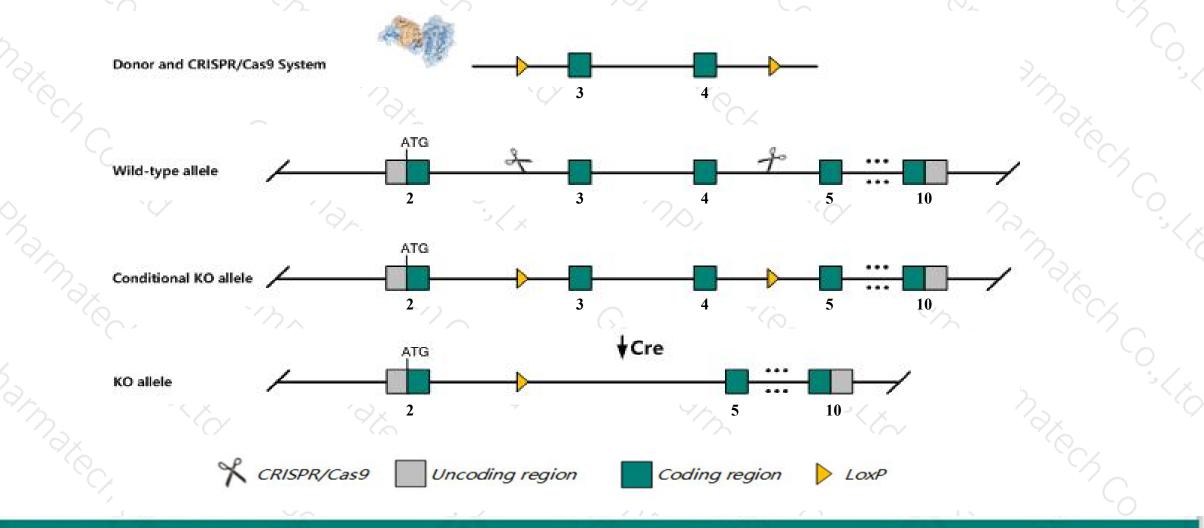


Conditional Knockout strategy



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This model will use CRISPR/Cas9 technology to edit the Gnb4 gene. The schematic diagram is as follows:



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The Gnb4 gene has 8 transcripts. According to the structure of Gnb4 gene, exon3-exon4 of Gnb4-205 (ENSMUST00000155737.7) transcript is recommended as the knockout region. The region contains 146bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Gnb4* 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.



- The Gnb4 gene is located on the Chr3. 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)



Gnb4 guanine nucleotide binding protein (G protein), beta 4 [Mus musculus (house mouse)]

Gene ID: 14696, updated on 24-Oct-2019

Summary

Official SymbolGnb4 provided by MGIOfficial Full Nameguanine nucleotide binding protein (G protein), beta 4 provided by MGIPrimary sourceMGI:MGI:104581See relatedEnsembl:ENSMUSG0000027669Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae;
Murinae; Mus; MusAlso known asG(beta)4; 6720453A21RikExpressionBroad expression in placenta adult (RPKM 17.2), CNS E18 (RPKM 11.9) and 23 other tissues See more
human all

Genomic context

☆ ?

See Gnb4 in Genome Data Viewer

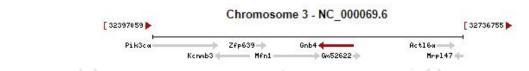
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Location: 3: 3 A3

2004110111 0,071

Exon count: 13

Annotation release	Status	Assembly	Chr	Location
<u>108</u>	current	GRCm38.p6 (GCF_000001635.26)	3	NC_000069.6 (3257858732616673, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	3	NC_000069.5 (3248245032515457, complement)



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Transcript information (Ensembl)



The gene has 8 transcripts, all transcripts are shown below:

Name 🍦	Transcript ID	bp 🖕	Protein 🖕	Biotype 🍦	CCDS 🍦	UniProt 🍦	Flags		
Gnb4-205	ENSMUST00000155737.7	5997	<u>340aa</u>	Protein coding	<u>CCDS17297</u> 교	P29387	TSL:1 GENCODE basic APPRIS P1		
Gnb4-201	ENSMUST00000108234.7	2510	<u>340aa</u>	Protein coding	<u>CCDS17297</u> 函	P29387	TSL:1 GENCODE basic APPRIS P1		
Gnb4-208	ENSMUST00000193050.1	423	<u>73aa</u>	Protein coding	8	<u>A0A0A6YVN9</u> &	TSL:5 GENCODE basic		
Gnb4-206	ENSMUST00000184130.6	1356	<u>83aa</u>	Nonsense mediated decay	8	<u>V9GWY1</u> 团	TSL:1		
Gnb4-203	ENSMUST00000144292.1	4395	No protein	Retained intron	8	18,55.8	TSL:1		
Gnb4-202	ENSMUST00000143588.1	2388	No protein	Retained intron	8	858	TSL:1		
Gnb4-204	ENSMUST00000152901.7	2357	No protein	Retained intron	8	858	TSL:1		
Gnb4-207	ENSMUST00000192116.1	728	No protein	Retained intron	8	8 .	TSL:3		

The strategy is based on the design of *Gnb4-205* transcript, The transcription is shown below

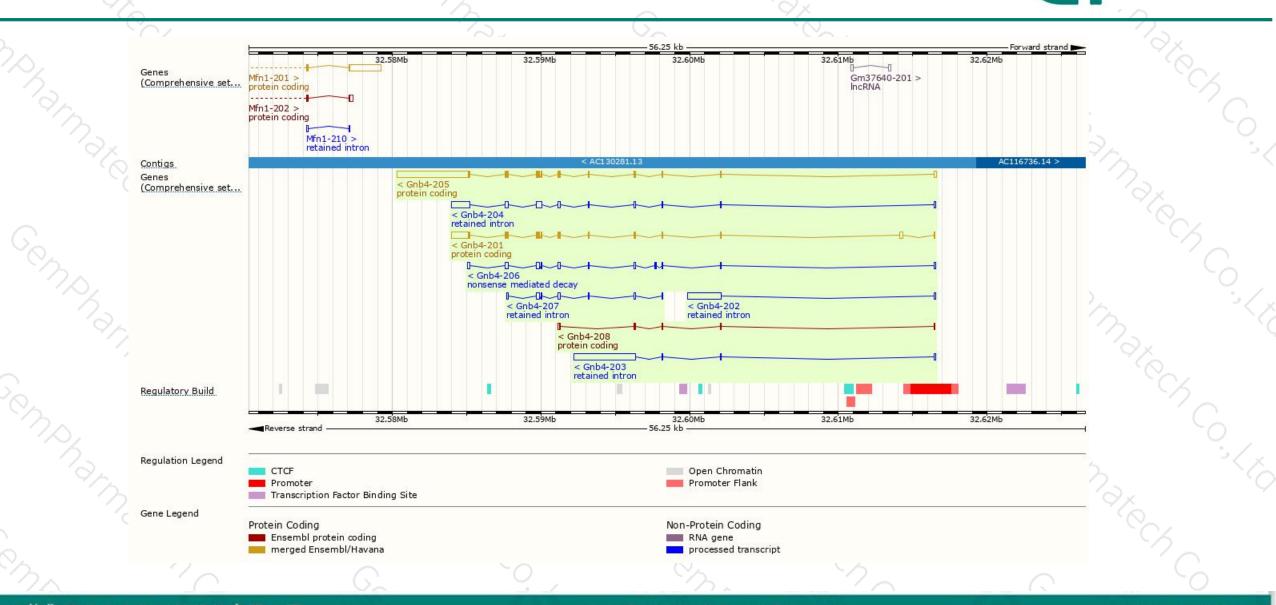
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< Gnb4-205

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Genomic location distribution



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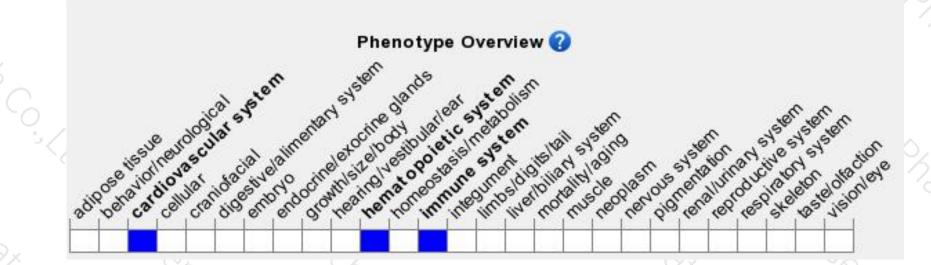
Protein domain



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- Pr	ENSMUSP00000121 Low.complexity (Seg) Coiled-coils (Ncoils)								
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	PIRSF	PIRSF002394	worke repeat, conser	ved site					
1/	PANTHER	Guanine nucleotide-binding prote	in, beta subunit						
		PTHR19850:SF28							-5/
	Gene3D	WD40/YVTN repeat-like-containir	ig domain superfamily						
	CDD		0200						
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# Mouse phenotype description(MGI)





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

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



