Gnao1 Cas9-KO Strategy

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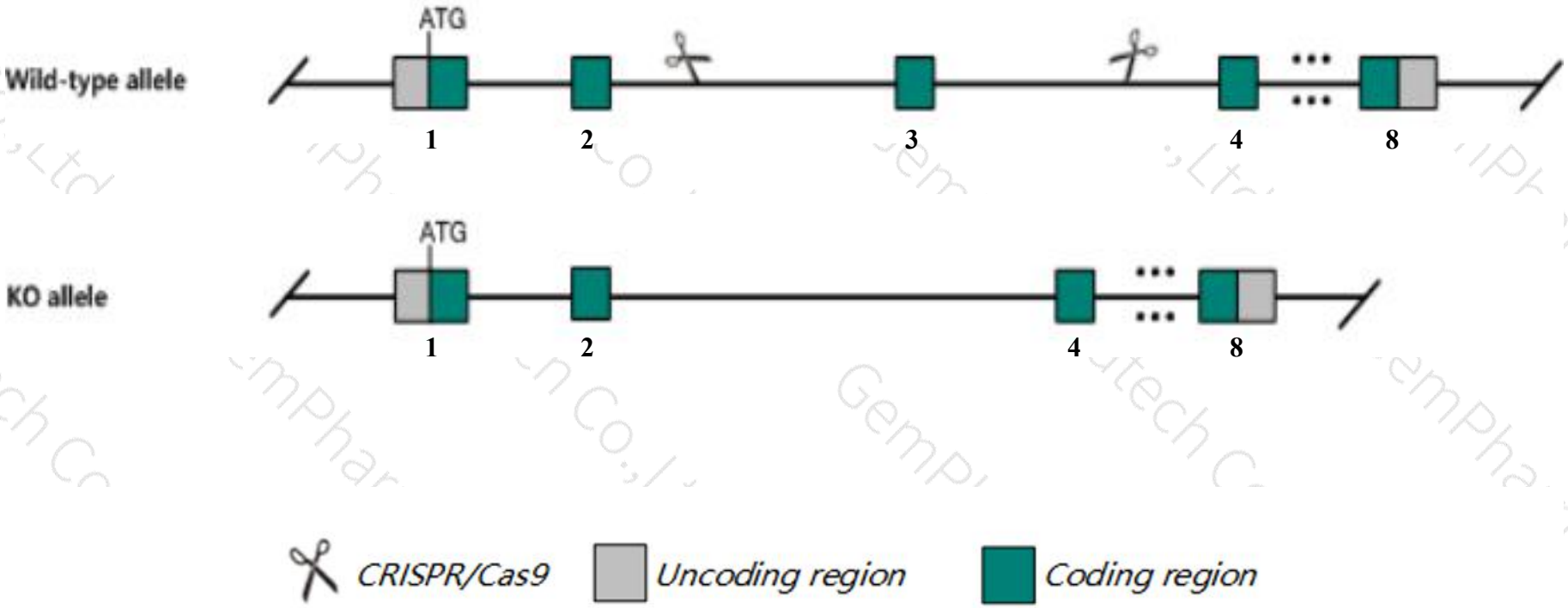
Project Overview

Project Name	***Gnao1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gnao1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Gnao1* gene has 12 transcripts. According to the structure of *Gnao1* gene, exon3 of *Gnao1-203*(ENSMUST00000125716.7) transcript is recommended as the knockout region.The region contains 142bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Gnao1* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ According to the existing MGI data,mice lacking both isoforms exhibit reduced survival, sterility, low body weight, hyperalgesia, tremors, turning behavior, impaired locomotion, altered channel response and improved glucose tolerance. Isoform-specific deletion may lead to increased insulin release and abnormal eye electrophysiology.

➢ The *Gnao1* gene is located on the Chr8. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

集萃药康 GemPharmatech

Gnao1 guanine nucleotide binding protein, alpha O [Mus musculus (house mouse)]

Gene ID: 14681, updated on 13-Mar-2020

Summary

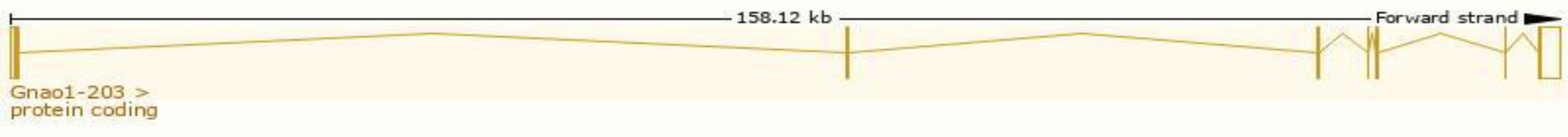
Official Symbol	Gnao1 provided by MGI
Official Full Name	guanine nucleotide binding protein, alpha O provided by MGI
Primary source	MGI:MGI:95775
See related	Ensembl:ENSMUSG00000031748
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AW050213, Galphao, Gnao, alphaO
Expression	Broad expression in cortex adult (RPKM 40.0), frontal lobe adult (RPKM 39.2) and 16 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

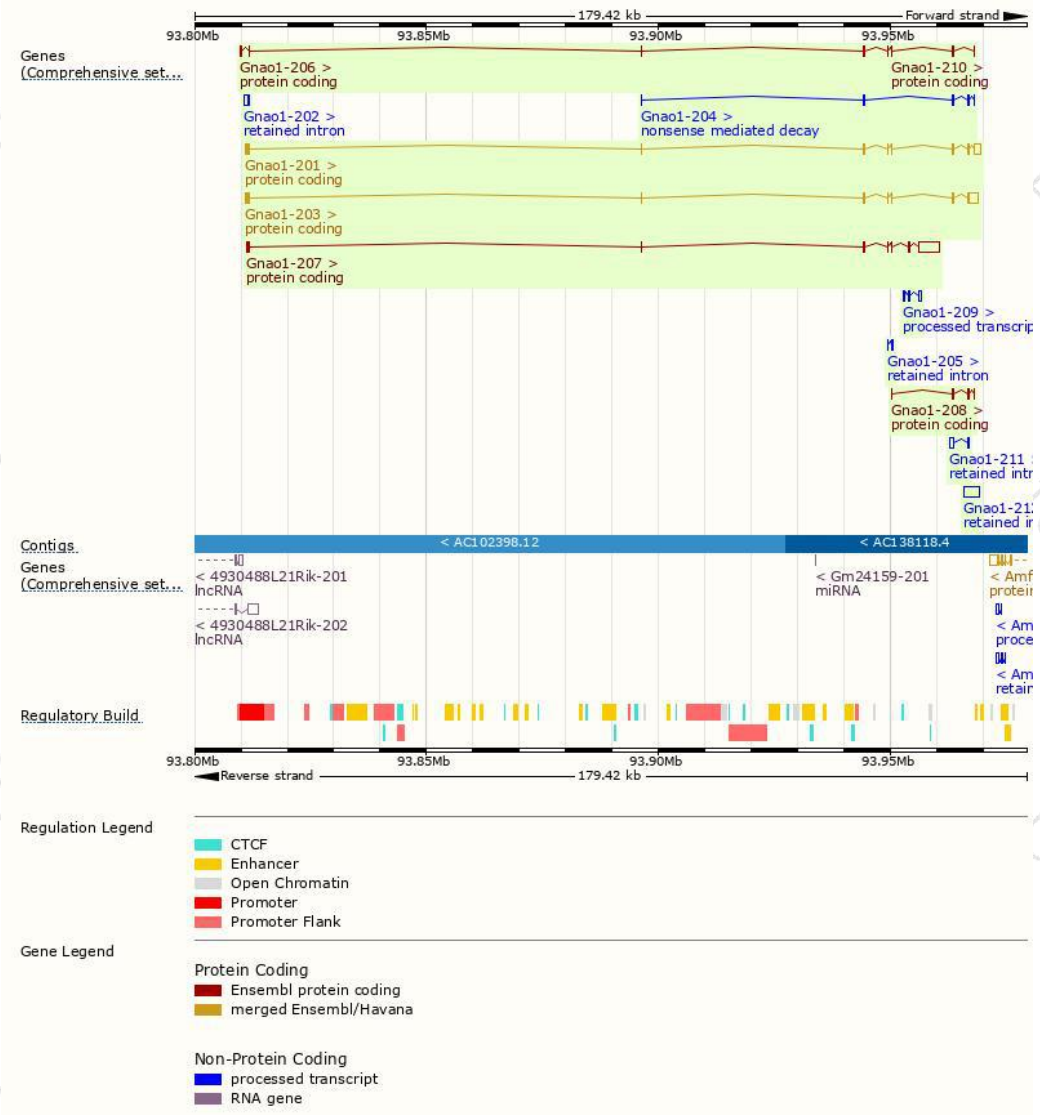
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnao1-207	ENSMUST00000138659.8	5526	354aa	Protein coding	CCDS85585	P18872	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Gnao1-203	ENSMUST00000125716.7	3462	354aa	Protein coding	CCDS22532	P18872 Q543S2	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Gnao1-201	ENSMUST00000034198.14	2948	354aa	Protein coding	CCDS22532	P18872 Q543S2	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Gnao1-206	ENSMUST00000137202.7	741	179aa	Protein coding	-	D3Z2M7	CDS 3' incomplete TSL:3
Gnao1-208	ENSMUST00000142466.1	332	109aa	Protein coding	-	F6WC15	CDS 5' incomplete TSL:5
Gnao1-210	ENSMUST00000149530.7	231	74aa	Protein coding	-	F6W1B2	CDS 5' incomplete TSL:5
Gnao1-204	ENSMUST00000127900.1	805	101aa	Nonsense mediated decay	-	F7BLT7	CDS 5' incomplete TSL:5
Gnao1-209	ENSMUST00000144451.1	748	No protein	Processed transcript	-	-	TSL:3
Gnao1-212	ENSMUST00000212008.1	3415	No protein	Retained intron	-	-	TSL:NA
Gnao1-202	ENSMUST00000125695.2	938	No protein	Retained intron	-	-	TSL:1
Gnao1-211	ENSMUST00000155245.1	848	No protein	Retained intron	-	-	TSL:2
Gnao1-205	ENSMUST00000130974.1	402	No protein	Retained intron	-	-	TSL:3

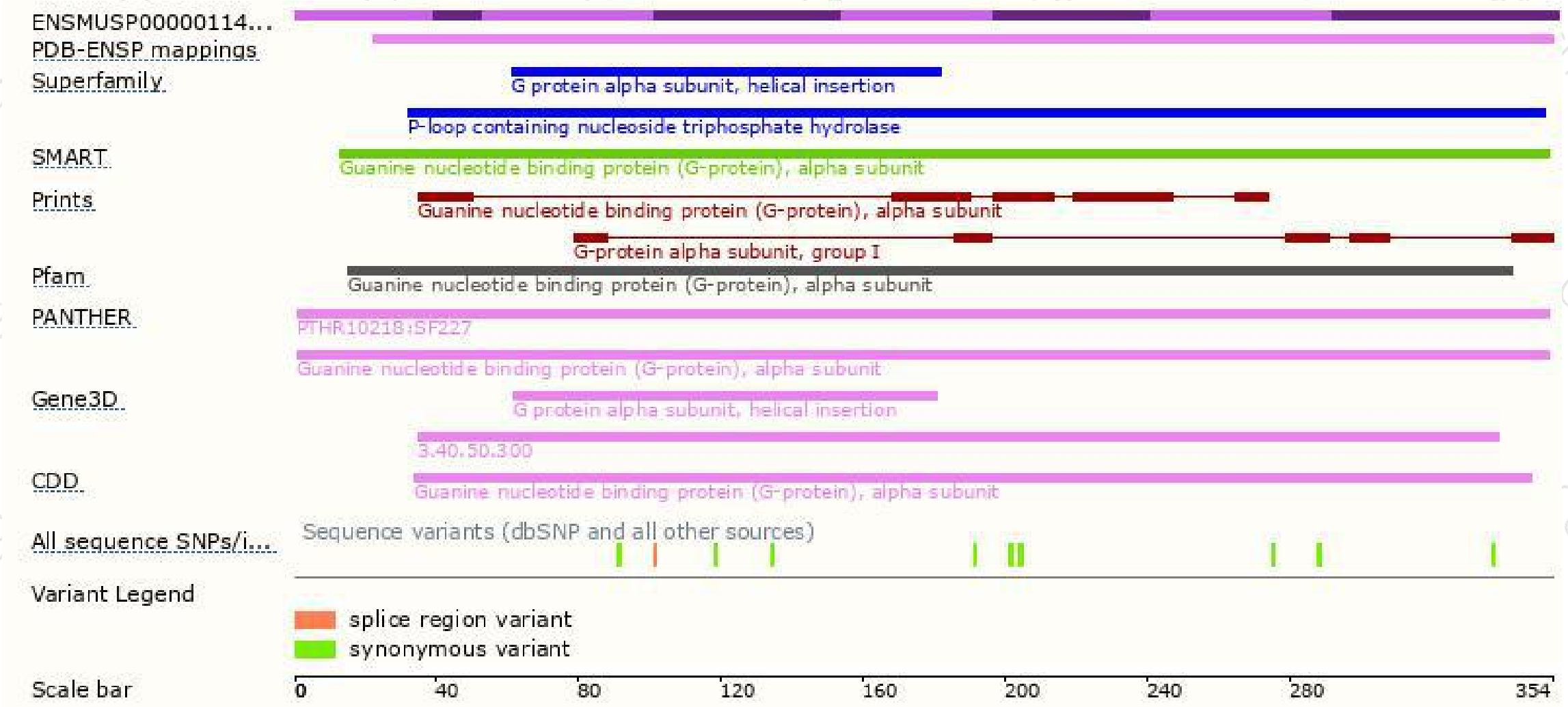
The strategy is based on the design of *Gnao1-203* transcript,the transcription is shown below:



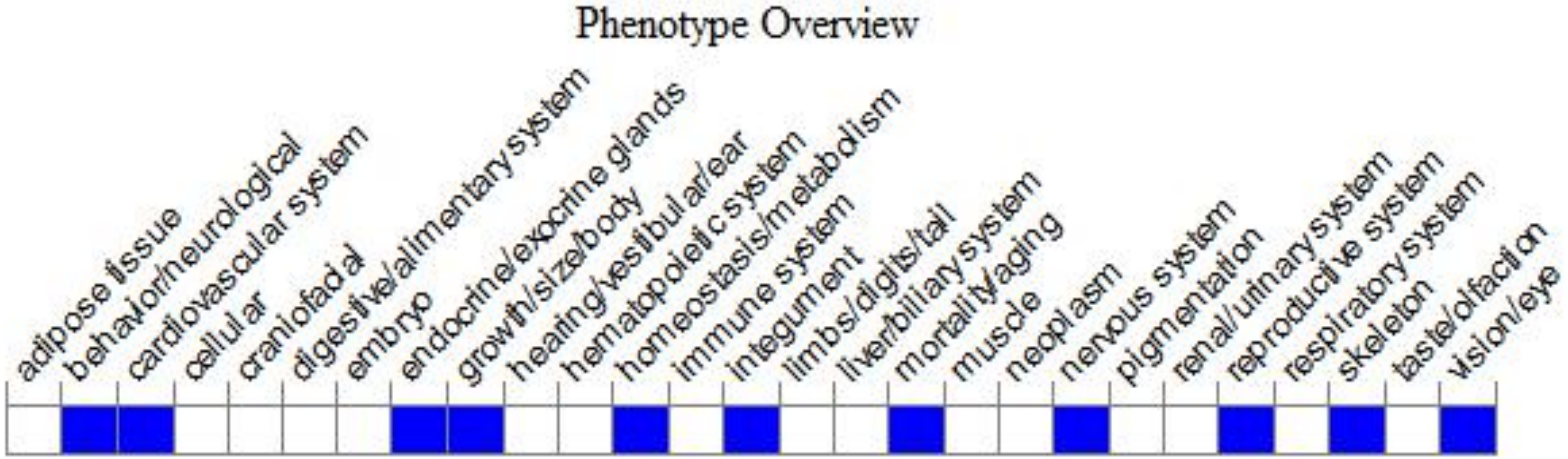
Genomic location distribution



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/).

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If you have any questions, you are welcome to inquire.

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