

# Gnaq Cas9-KO Strategy

Designer: Daohua Xu

Reviewer: Huimin Su

Design Date: 2023-1-16

## Overview

### Target Gene Name

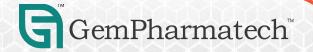
• Gnaq

### Project Type

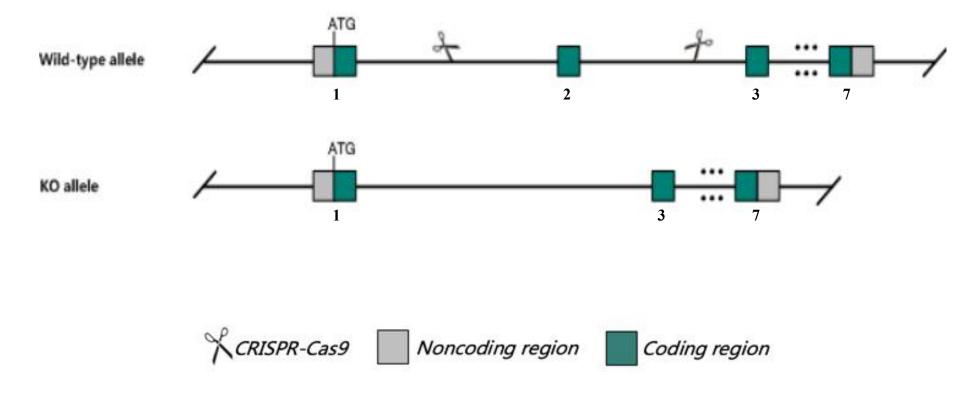
• Cas9-KO

### Genetic Background

• C57BL/6JGpt



## Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *Gnaq* gene.



### Technical Information

- The *Gnaq* gene has 3 transcripts. According to the structure of *Gnaq* gene, exon 2 of *Gnaq-201* (ENSMUST00000025541.6) transcript is recommended as the knockout region. The region contains 185bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Gnaq* gene. The brief process is as follows: gRNAs were transcribed in vitro. Cas9 and gRNAs 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.



### Gene Information

#### Gnaq guanine nucleotide binding protein, alpha q polypeptide [Mus musculus (house mouse)]

Gene ID: 14682, updated on 12-Jul-2022

#### Summary

☆ ?

Official Symbol Gnaq provided by MGI

Official Full Name guanine nucleotide binding protein, alpha q polypeptide provided by MGI

Primary source MGI:MGI:95776

See related Ensembl: ENSMUSG00000024639

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 1110005L02Rik, 6230401l02Rik, AA408290, AW060788, Dsk1, Dsk10, Galphaq, Gq, Gql

Expression Ubiquitous expression in CNS E18 (RPKM 21.9), CNS E14 (RPKM 19.1) and 28 other tissuesSee more

Orthologs <u>human all</u>

Source: https://www.ncbi.nlm.nih.gov/

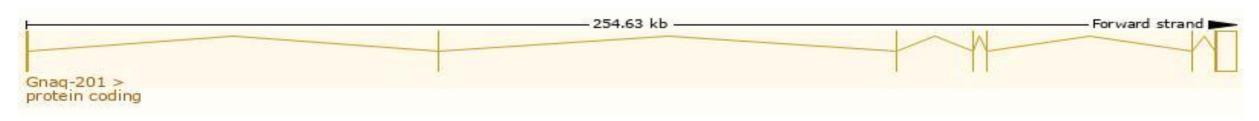


## Transcript Information

The gene has 3 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnaq-201	ENSMUST00000025541.6	5644	359aa	Protein coding	CCDS29684		TSL:1, GENCODE basic, APPRIS P1,
Gnaq-202	ENSMUST00000167656.2	481	No protein	Processed transcript	100		TSL:3,
Gnaq-203	ENSMUST00000170229.2	862	No protein	Retained intron	72		TSL:2,

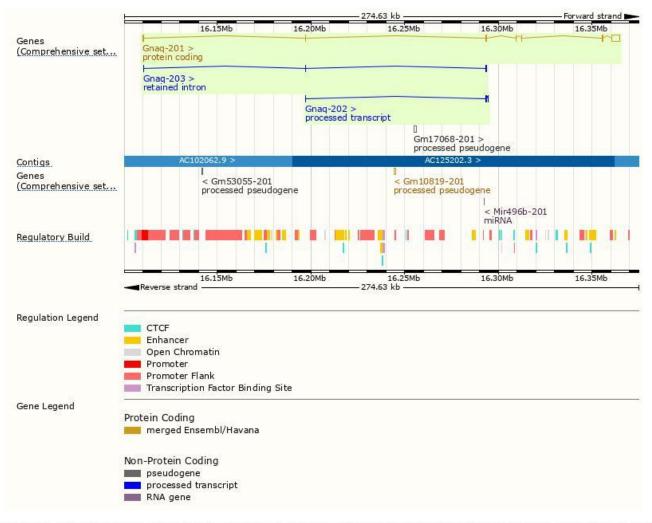
The strategy is based on the design of *Gnaq-201* transcript, the transcription is shown below:



Source: https://www.ensembl.org



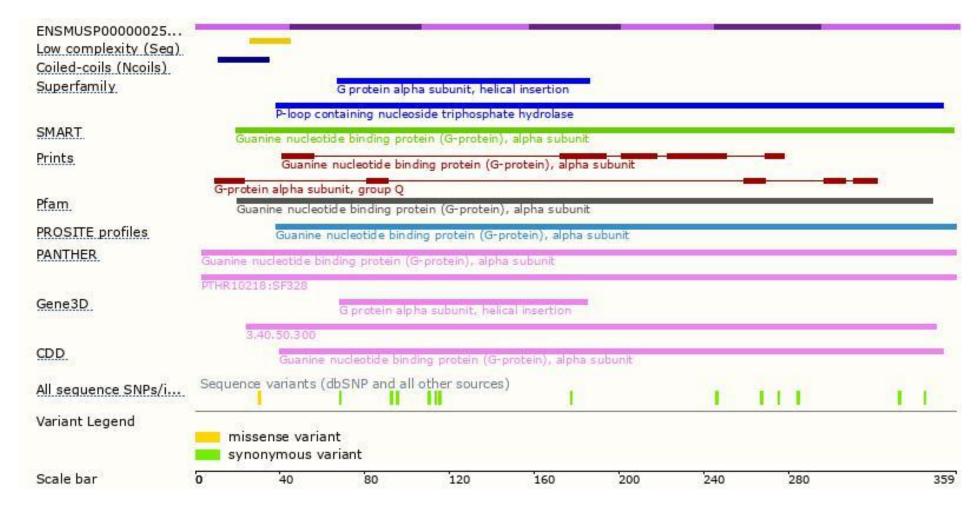
## Genomic Information





Source: : https://www.ensembl.org

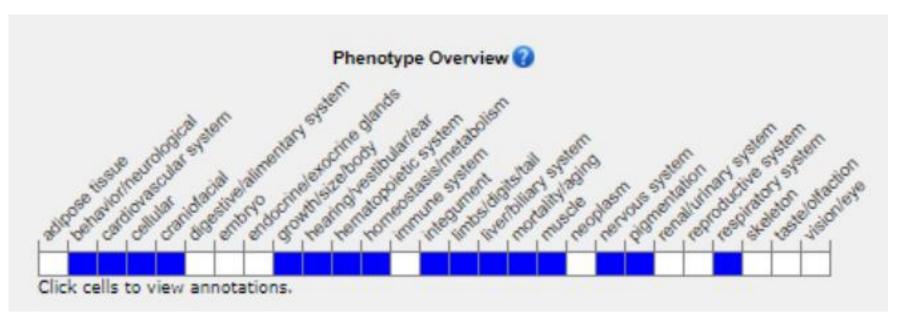
## Protein Information





Source: : https://www.ensembl.org

## Mouse Phenotype Information (MGI)



• Mutant mice exhibit pigmentation anomalies affecting the ears, tail and footpads.



## Important Information

- According to the existing MGI data, 30% 40% knockout homozygous mice died after birth.
- *Gnaq* is located on Chr19. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risks of the mutation on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

