

Amigo2 Cas9-KO Strategy

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Design Date: 2024-4-8

Overview

Target Gene Name

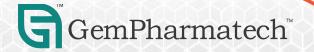
• Amigo2

Project Type

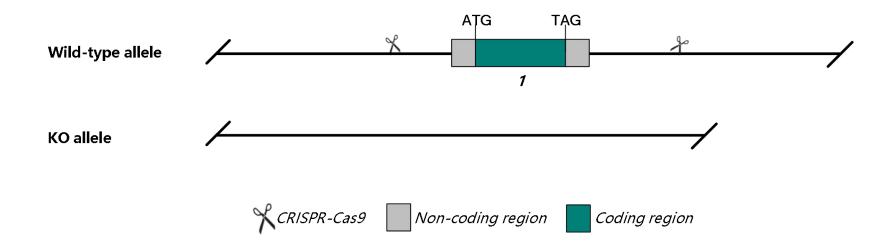
• Cas9-KO

Genetic Background

• C57BL/6JGpt



Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the Amigo2 gene.



Technical Information

- The *Amigo2* gene has 2 transcripts. According to the structure of *Amigo2* gene, exon 1 of *Amigo2*-202 (ENSMUST00000229890.2) is recommended as the knockout region. The region contains all of coding sequence. Knocking out the region will result in disruption of gene function.
- In this project we use CRISPR-Cas9 technology to modify *Amigo2* 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 ontarget 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

Amigo2 adhesion molecule with Ig like domain 2 [Mus musculus (house mouse)]

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Gene ID: 105827, updated on 5-Mar-2024



Genomic context

↑ ?

Location: 15 F1; 15 52.91 cM

See Amigo2 in Genome Data Viewer

Exon count: 2

https://www.ncbi.nlm.nih.gov/gene/105827

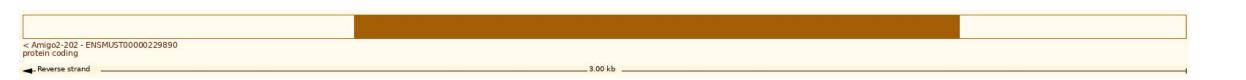


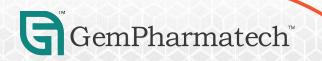
Transcript Information

The gene has 2 transcripts, all transcripts are shown below:

| Show/hide columns (1 hidden) | | | | | | | | Filter | | |
|------------------------------|------------|------|--------------|----------------|-------------|-------------------|-------------------|----------------|-----------|--|
| Transcript ID 👙 | Name ▼ | bp 👙 | Protein 🍦 | Biotype | CCDS 🍦 | UniProt Match | Flags | | | |
| ENSMUST00000229890.2 | Amigo2-202 | 2998 | <u>519aa</u> | Protein coding | CCDS27780@ | Q4VBE6 ₽ Q80ZD9 ₽ | Ensembl Canonical | GENCODE basic | APPRIS P1 | |
| ENSMUST00000053106.7 | Amigo2-201 | 2801 | <u>519aa</u> | Protein coding | CCDS27780 ₽ | Q4VBE6광 Q80ZD9광 | GENCODE b | asic APPRIS P1 | TSL:1 | |

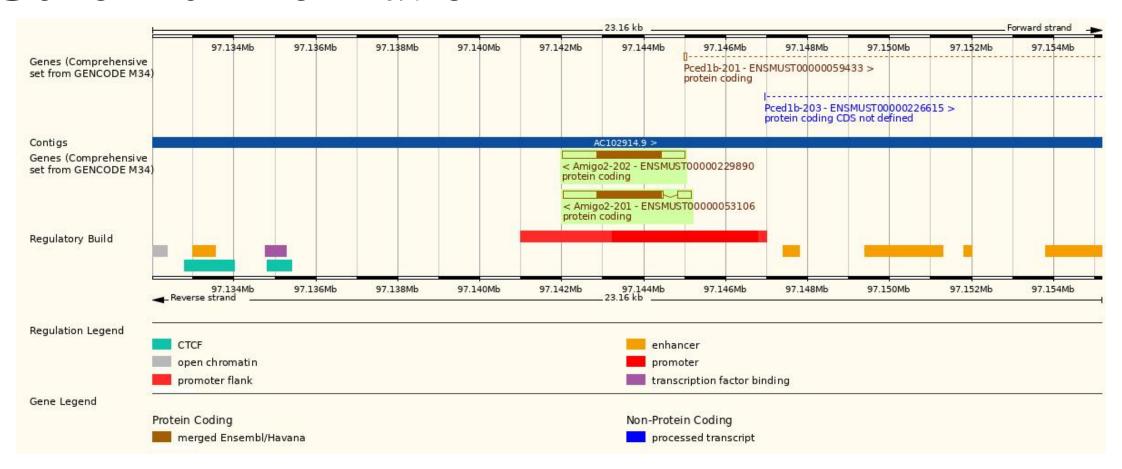
The strategy is based on the design of *Amigo2*-202 transcript, the transcription is shown below:





Source: http://asia.ensembl.org/

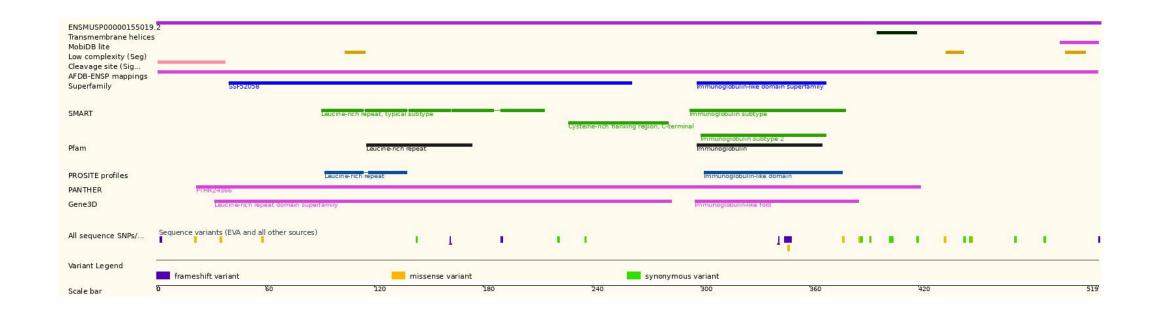
Genomic Information





Source: http://asia.ensembl.org/

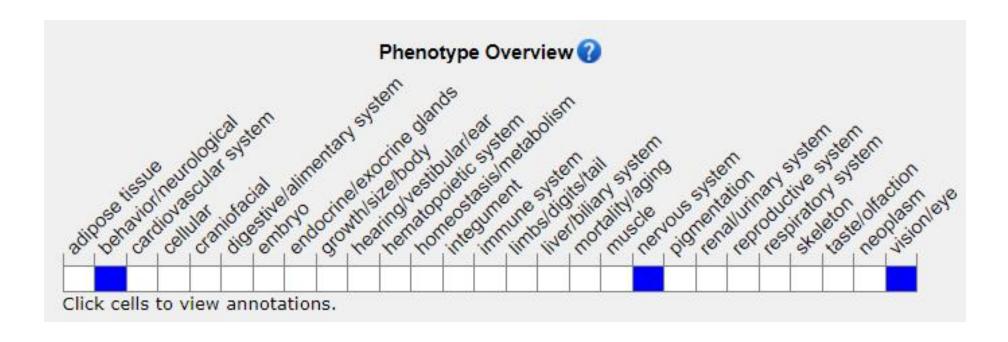
Protein Information





Source: https://www.ensembl.org

Mouse Phenotype Information (MGI)



Homozygous null mice exhibit starburst amacrine cell and rod bipolar cell dendrite arbor expansion and enhanced direction selectivity of direction-selective ganglion cell responses to starburst amacrine cell signals.



Source: https://www.informatics.jax.org

Important Information

- The knockout region overlaps with *Pced1b* gene, which may affect the function of this gene.
- *Amigo2* is located on Chr 15. 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 risk on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

