Ngef Cas9-KO Strategy

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Project Overview

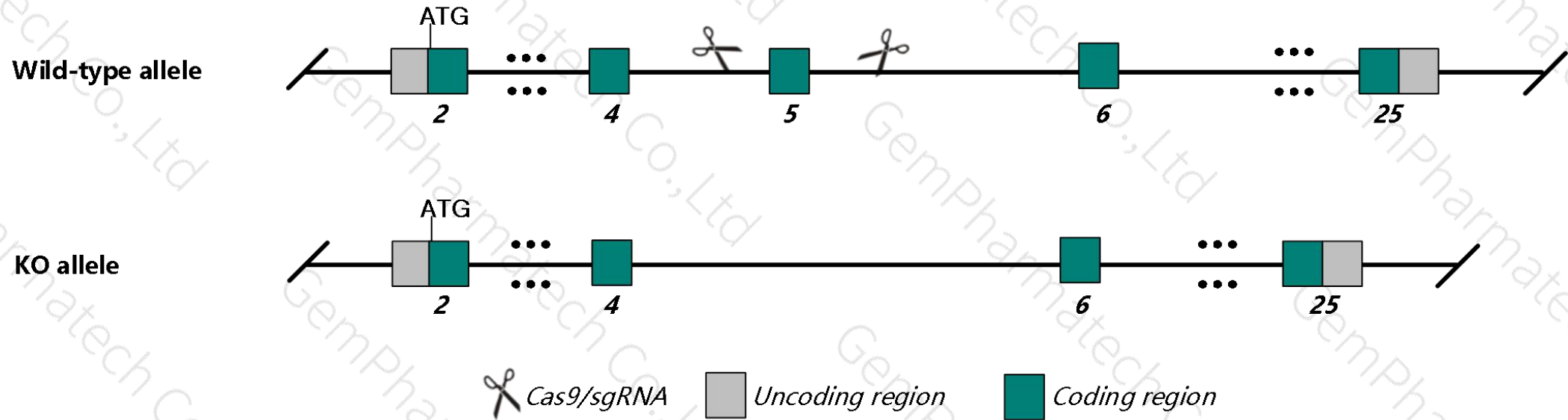|	
Project Name	*Ngef*
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

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

Technical routes

➢ The *Ngef* gene has 4 transcripts. According to the structure of *Ngef* gene, exon5 of *Ngef-202* (ENSMUST00000068681.11) transcript is recommended as the knockout region.The region contains 308bp coding sequence. Knock out the region will result in disruption of protein function.

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

Notice

➢ According to the MGI date, mice homozygous for a knock-out allele are viable and fertile and show no overt axonal phenotype; however, cultured retinal ganglion cells display defects in axonal outgrowth and ephrin-induced growth cone collapse.

➢ The transcript *Ngef-204* is incomplete, so the effect on it is unknown.

➢ The *Ngef* gene is located on the Chr1. 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）

Ngef neuronal guanine nucleotide exchange factor [*Mus musculus* (house mouse)]

Gene ID: 53972, updated on 13-Mar-2020

▲ Summary

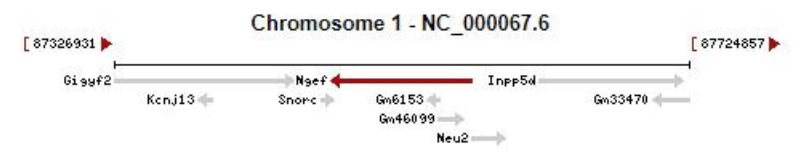|	
Official Symbol	Ngef provided by MGI
Official Full Name	neuronal guanine nucleotide exchange factor provided by MGI
Primary source	MGI:MGI:1858414
See related	Ensembl:ENSMUSG00000026259
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Tims2; ephexin
Expression	Biased expression in cortex adult (RPKM 59.5), frontal lobe adult (RPKM 38.3) and 10 other tissues See more
Orthologs	human all

▲ Genomic context

Location: 1 D; 1 44.42 cM

See Ngef in Genome Data Viewer

Exon count: 16

| Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	1	NC_000067.6 (87476829..87573870, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	1	NC_000067.5 (89373404..89470445, complement)

Chromosome 1 - NC_000067.6

[87326931 ▶   [87724857 ▶

Gigyf2   Ngef   Inpp5d
Kcnj13   Snorc   Gm6153   Gm33470
Gm46099
Neu2

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Transcript information（Ensembl）

The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags		
Ngef-202	ENSMUST00000068681.11	3109	710aa	Protein coding	CCDS48307	E9QK62	TSL:1	GENCODE basic	APPRIS P4
Ngef-201	ENSMUST00000027477.14	2746	620aa	Protein coding	CCDS48306	Q8CHT1	TSL:1	GENCODE basic	APPRIS ALT2
Ngef-204	ENSMUST00000168235.1	1110	245aa	Protein coding	-	F6SWQ9		CDS 5' incomplete	TSL:3
Ngef-203	ENSMUST00000166463.1	735	No protein	Retained intron	-	-		TSL:2	

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

< Ngef-202
protein coding

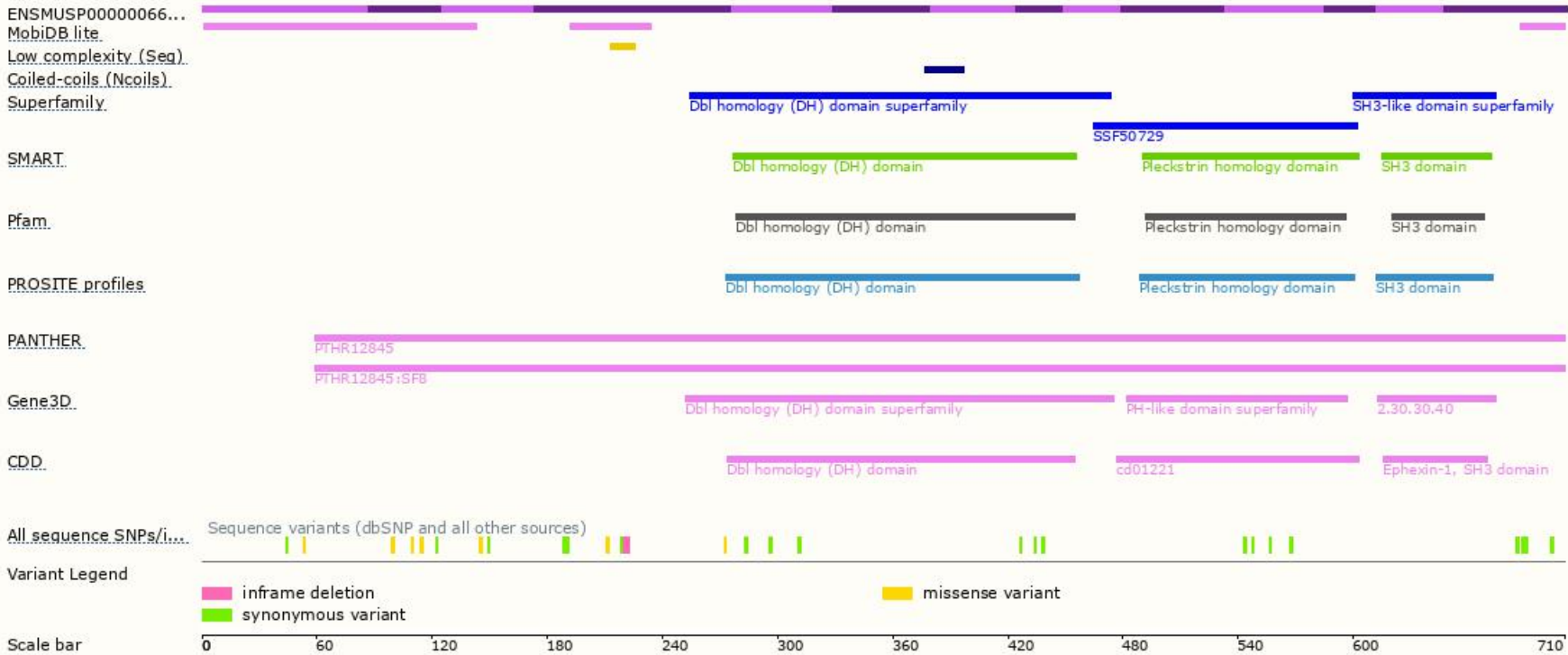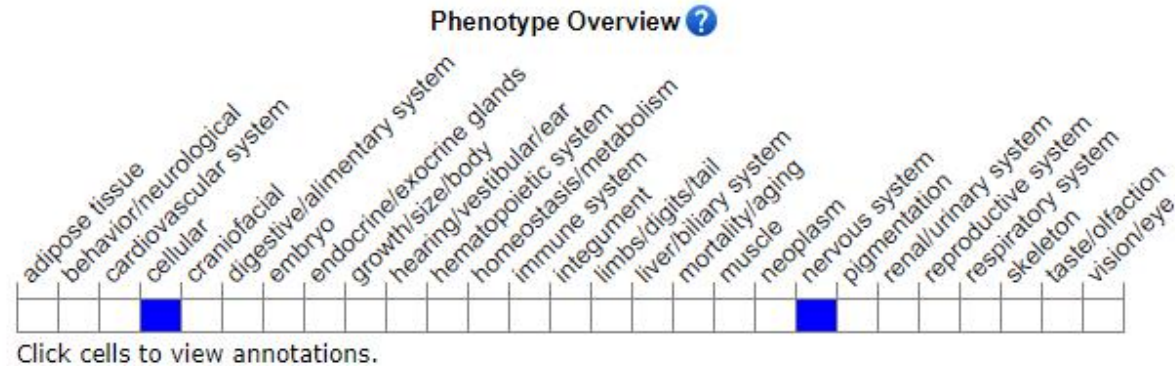◄ Reverse strand ──────────────── 97.04 kb ────────────────
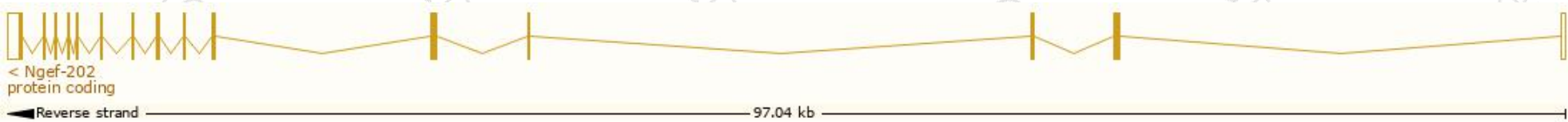
Genomic location distribution
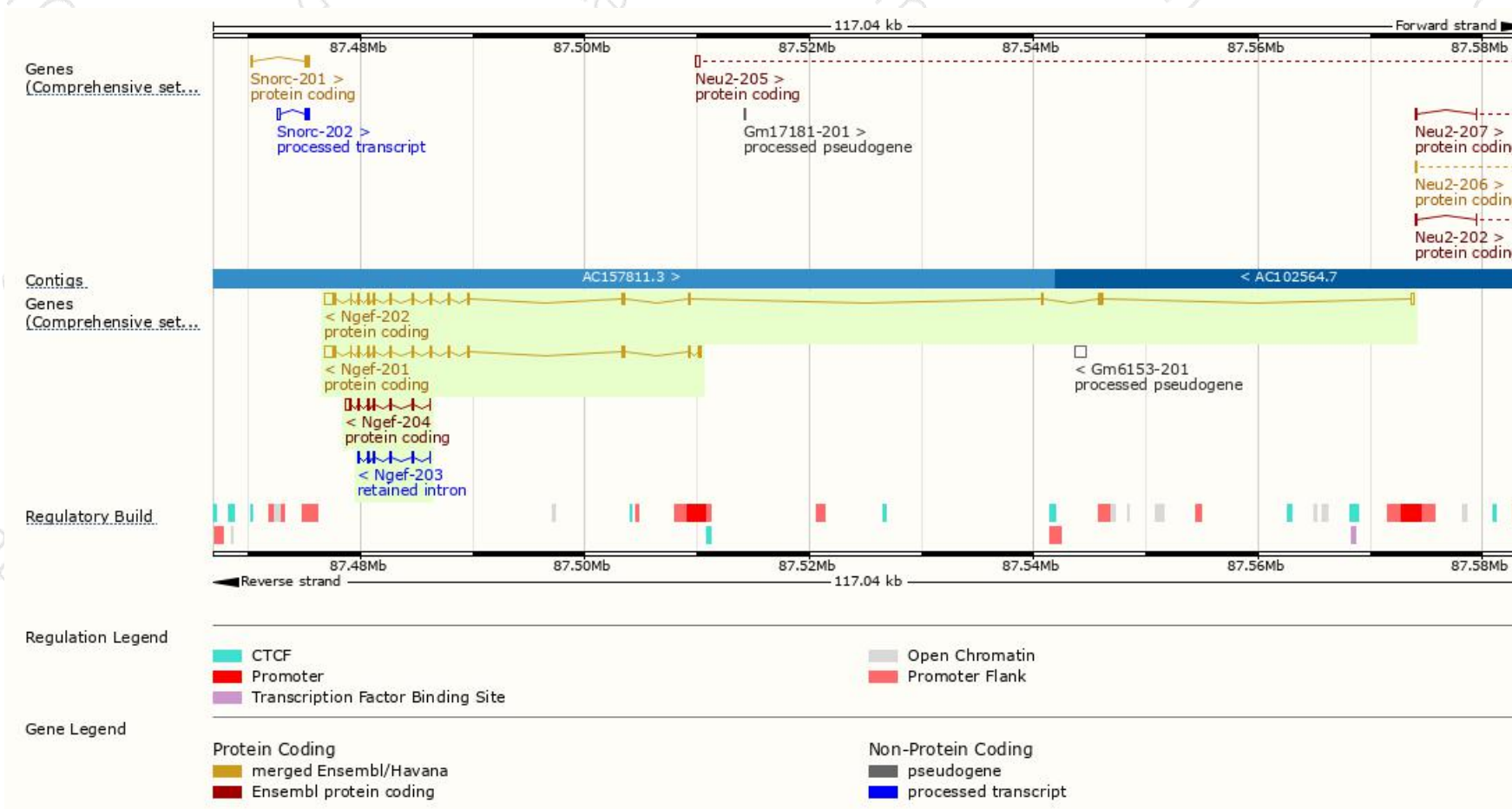
Protein domain

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, mice homozygous for a knock-out allele are viable and fertile and show no overt axonal phenotype; however, cultured retinal ganglion cells display defects in axonal outgrowth and ephrin-induced growth cone collapse.

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