

# ***Neu2 Cas9-CKO Strategy***

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**Reviewer:**

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# Project Overview

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**Project Name**

*Neu2*

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**Project type**

**Cas9-CKO**

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**Strain background**

**C57BL/6JGpt**

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# Conditional Knockout strategy

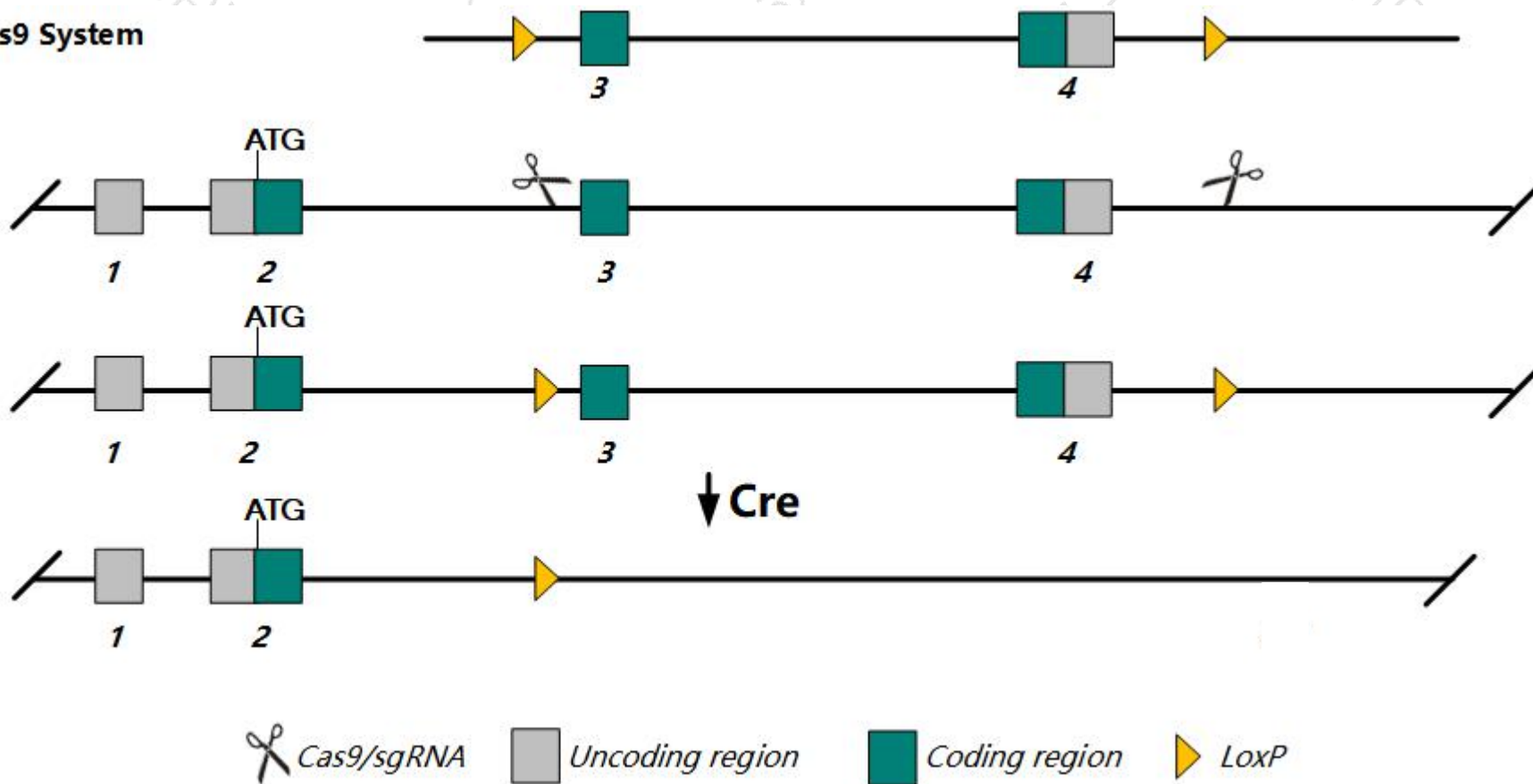
This model will use CRISPR/Cas9 technology to edit the *Neu2* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System

Wild-type allele

Conditional KO allele

KO allele



- The *Neu2* gene has 7 transcripts. According to the structure of *Neu2* gene, exon3-4 of *Neu2*-207 (ENSMUST00000172222.7) transcript is recommended as the knockout region. The region contains most coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Neu2* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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 or cell types.

# Notice

- The *Neu2* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.



# Gene information ( NCBI )

## Neu2 neuraminidase 2 [ *Mus musculus* (house mouse) ]

Gene ID: 23956, updated on 13-Mar-2020

### Summary



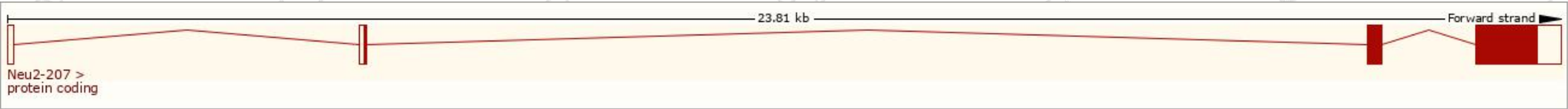
Official Symbol	Neu2 provided by <a href="#">MGI</a>
Official Full Name	neuraminidase 2 provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:1344417</a>
See related	<a href="#">Ensembl:ENSMUSG00000079434</a>
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	MBS; MSS; MTS
Expression	Biased expression in mammary gland adult (RPKM 11.9), kidney adult (RPKM 6.1) and 10 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

# Transcript information ( Ensembl )

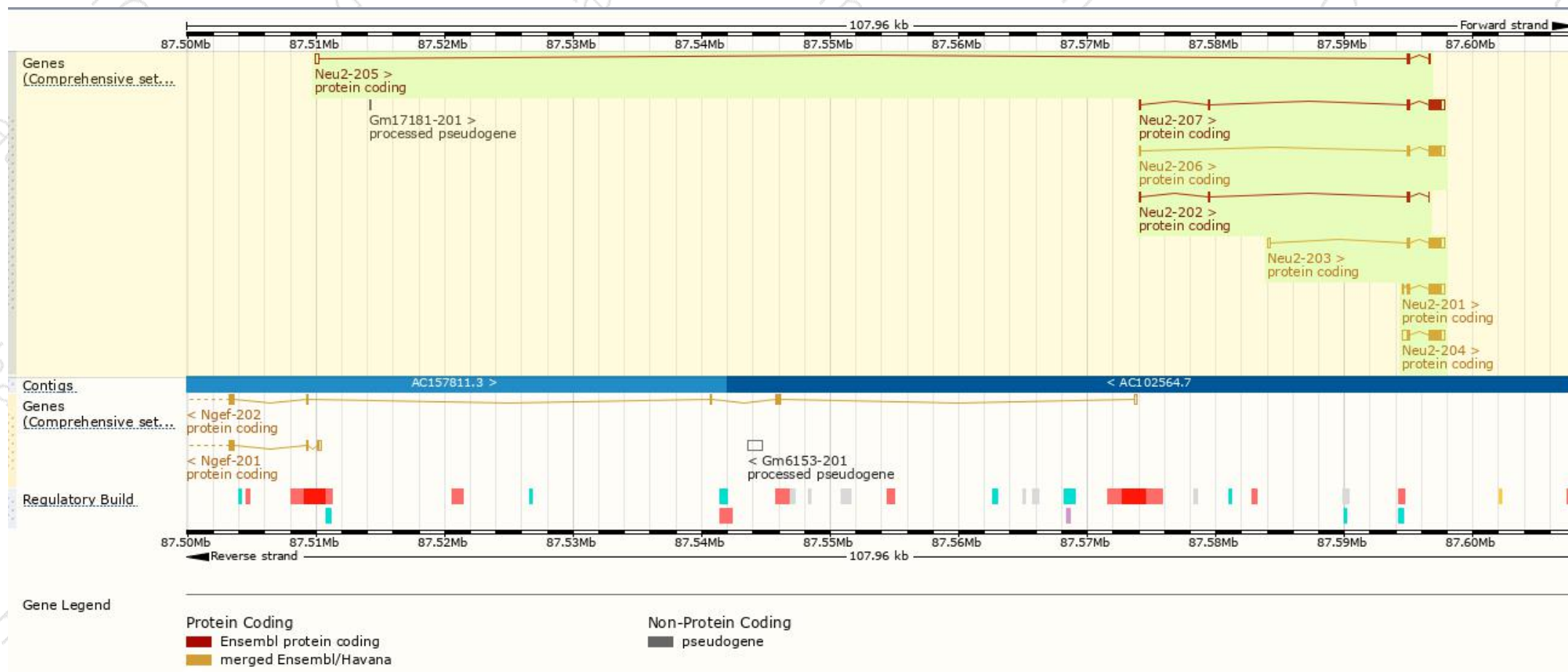
The gene has 7 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Neu2-204	<a href="#">ENSMUST00000165109.1</a>	1824	<a href="#">379aa</a>	Protein coding	<a href="#">CCDS15135</a>	<a href="#">Q0VGI4</a> <a href="#">Q9JMH3</a>	TSL:1 GENCODE basic APPRIS P3
Neu2-203	<a href="#">ENSMUST00000164128.1</a>	1733	<a href="#">385aa</a>	Protein coding	<a href="#">CCDS48309</a>	<a href="#">A0A0R4J224</a>	TSL:1 GENCODE basic APPRIS ALT2
Neu2-207	<a href="#">ENSMUST00000172222.7</a>	1715	<a href="#">393aa</a>	Protein coding	<a href="#">CCDS48308</a>	<a href="#">E9Q0I0</a>	TSL:1 GENCODE basic APPRIS ALT2
Neu2-206	<a href="#">ENSMUST00000166259.7</a>	1596	<a href="#">379aa</a>	Protein coding	<a href="#">CCDS15135</a>	<a href="#">Q0VGI4</a> <a href="#">Q9JMH3</a>	TSL:1 GENCODE basic APPRIS P3
Neu2-201	<a href="#">ENSMUST00000070898.5</a>	1570	<a href="#">379aa</a>	Protein coding	<a href="#">CCDS15135</a>	<a href="#">Q0VGI4</a> <a href="#">Q9JMH3</a>	TSL:1 GENCODE basic APPRIS P3
Neu2-205	<a href="#">ENSMUST00000166055.7</a>	672	<a href="#">111aa</a>	Protein coding	-	<a href="#">E9Q2Z0</a>	CDS 3' incomplete TSL:3
Neu2-202	<a href="#">ENSMUST00000163606.7</a>	423	<a href="#">90aa</a>	Protein coding	-	<a href="#">E9Q0P5</a>	CDS 3' incomplete TSL:1

The strategy is based on the design of *Neu2-207* transcript, The transcription is shown below



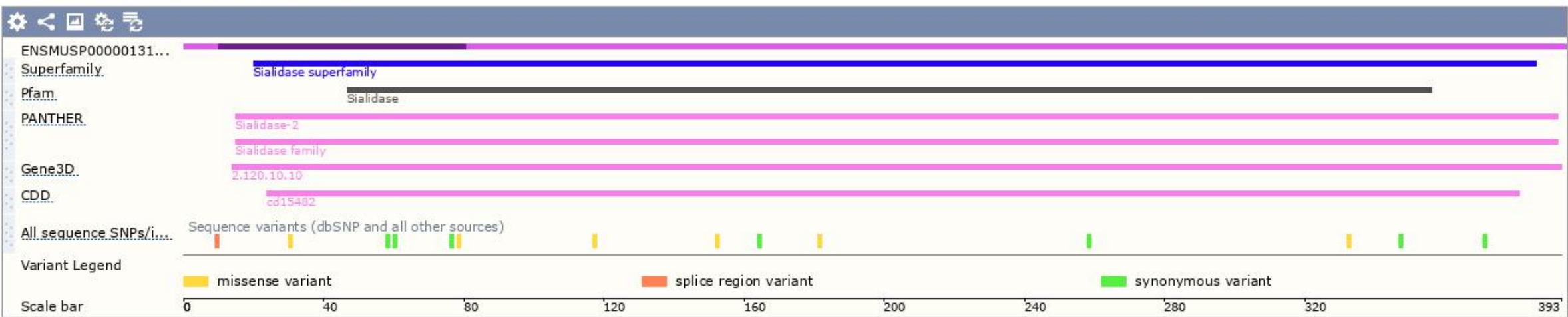
# Genomic location distribution





# Protein domain

Protein domains for ENSMUSP00000131409.1



Statistics

Ave. residue weight: 112.077 g/mol

Charge: 9.5

Isoelectric point: 7.6954

Molecular weight: 44,046.21 g/mol

Number of residues: 393 aa

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