Rhot1 Cas9-KO Strategy

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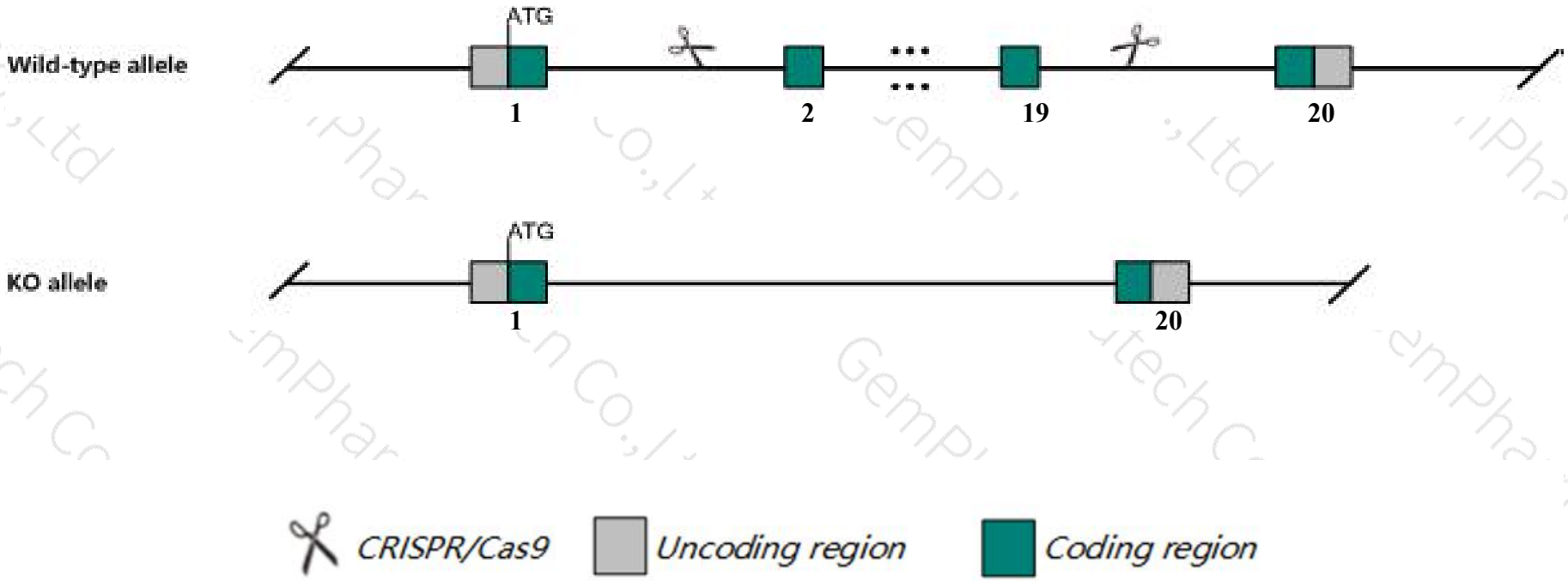
Project Overview

Project Name	***Rhot1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rhot1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Rhot1* gene has 11 transcripts. According to the structure of *Rhot1* gene, exon2-exon19 of *Rhot1-202* (ENSMUST00000055056.15) transcript is recommended as the knockout region.The region contains 1825bp coding sequenc Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Rhot1* gene. The brief process is as follows: CRISPR/Cas9 system

Notice

➢ According to the existing MGI data,Mice homozygous for a null allele die neonatally exhibiting cyanosis, respiratory failure, loss of brainstem cranial motor neurons, decreased cervical motor neuron number and phrenic nerve branching, and alterations in retrograde mitochondrial transport and run length in cortical axons.

➢Transcript 210 is unaffected.

➢ The *Rhot1* gene is located on the Chr11. 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）

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Rhot1 ras homolog family member T1 [*Mus musculus* (house mouse)]

Gene ID: 59040, updated on 21-Sep-2019

Summary

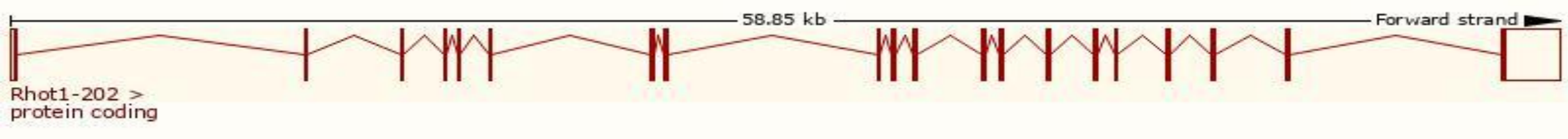
Official Symbol	Rhot1 provided by MGI
Official Full Name	ras homolog family member T1 provided by MGI
Primary source	MGI:MGI:1926078
See related	Ensembl:ENSMUSG00000017686
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Arht1; Miro1; AA415293; AF244542; AI834919; 2210403N23Rik; C430039G08Rik
Expression	Ubiquitous expression in CNS E18 (RPKM 10.8), bladder adult (RPKM 10.1) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

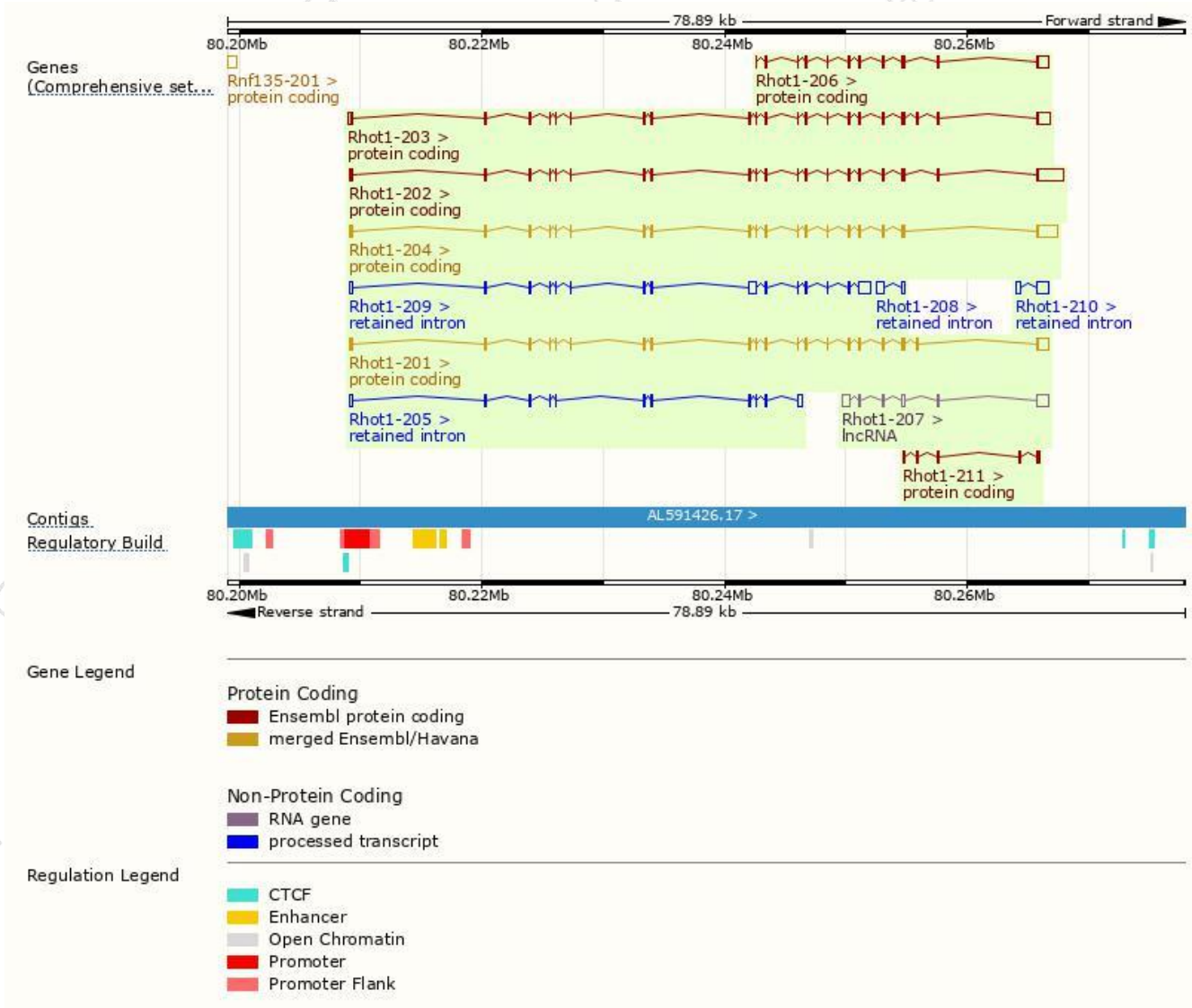
The gene has 11 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rhot1-202	ENSMUST00000055056.15	4260	672aa	Protein coding	CCDS48862	Q8BG51	TSL:1 GENCODE basic APPRIS ALT1
Rhot1-204	ENSMUST00000092857.12	3687	631aa	Protein coding	CCDS25130	Q8BG51	TSL:1 GENCODE basic APPRIS P3
Rhot1-201	ENSMUST00000017831.15	3029	663aa	Protein coding	CCDS48861	Q8BG51	TSL:1 GENCODE basic APPRIS ALT1
Rhot1-203	ENSMUST00000077451.13	3297	704aa	Protein coding	-	Q8BG51	TSL:5 GENCODE basic APPRIS ALT1
Rhot1-206	ENSMUST00000134894.1	2188	434aa	Protein coding	-	F7ASU3	CDS 5' incomplete TSL:1
Rhot1-211	ENSMUST00000237515.1	825	214aa	Protein coding	-	-	CDS 5' incomplete
Rhot1-209	ENSMUST00000154362.7	2954	No protein	Retained intron	-	-	TSL:1
Rhot1-210	ENSMUST00000236882.1	1347	No protein	Retained intron	-	-	
Rhot1-205	ENSMUST00000134148.1	1326	No protein	Retained intron	-	-	TSL:1
Rhot1-208	ENSMUST00000151518.1	759	No protein	Retained intron	-	-	TSL:2
Rhot1-207	ENSMUST00000135947.1	2192	No protein	lncRNA	-	-	TSL:1

The strategy is based on the design of *Rhot1-202* transcript,The transcription is shown below

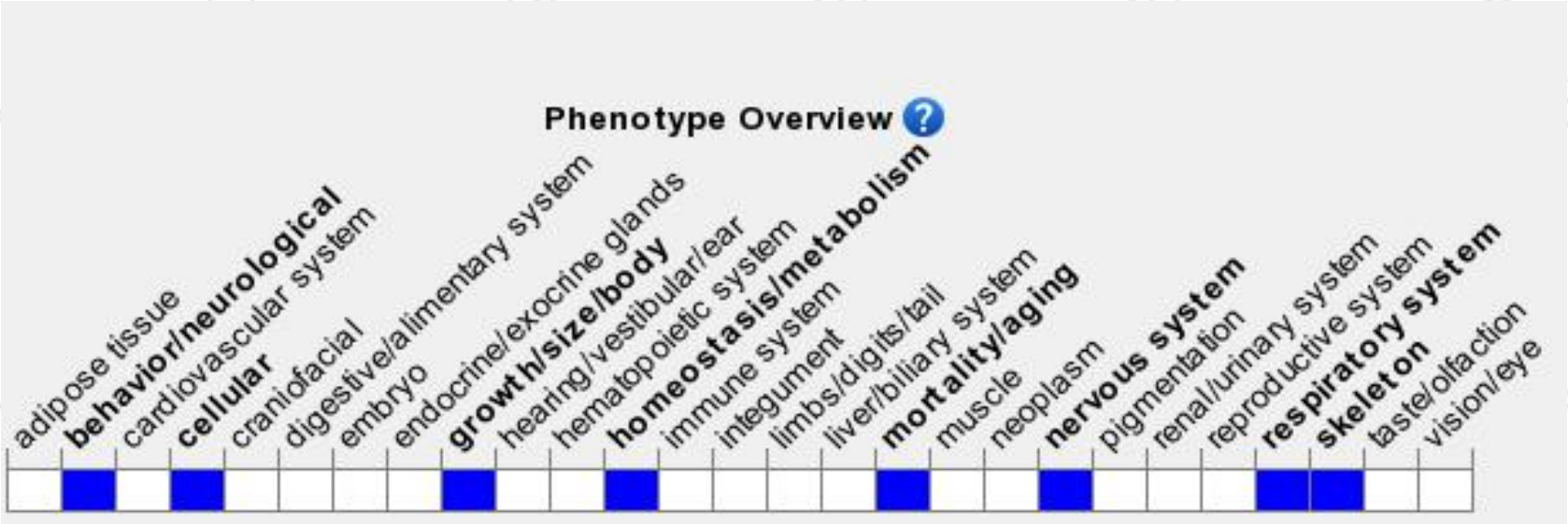


Genomic location distribution



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/).

According to the existing MGI data,Mice homozygous for a null allele die neonatally exhibiting cyanosis, respiratory failure, loss of brainstem cranial motor neurons, decreased cervical motor neuron number and phrenic nerve branching, and alterations in retrograde mitochondrial transport and run length in cortical axons.

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

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