Ncald Cas9-CKO Strategy

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Project Overview

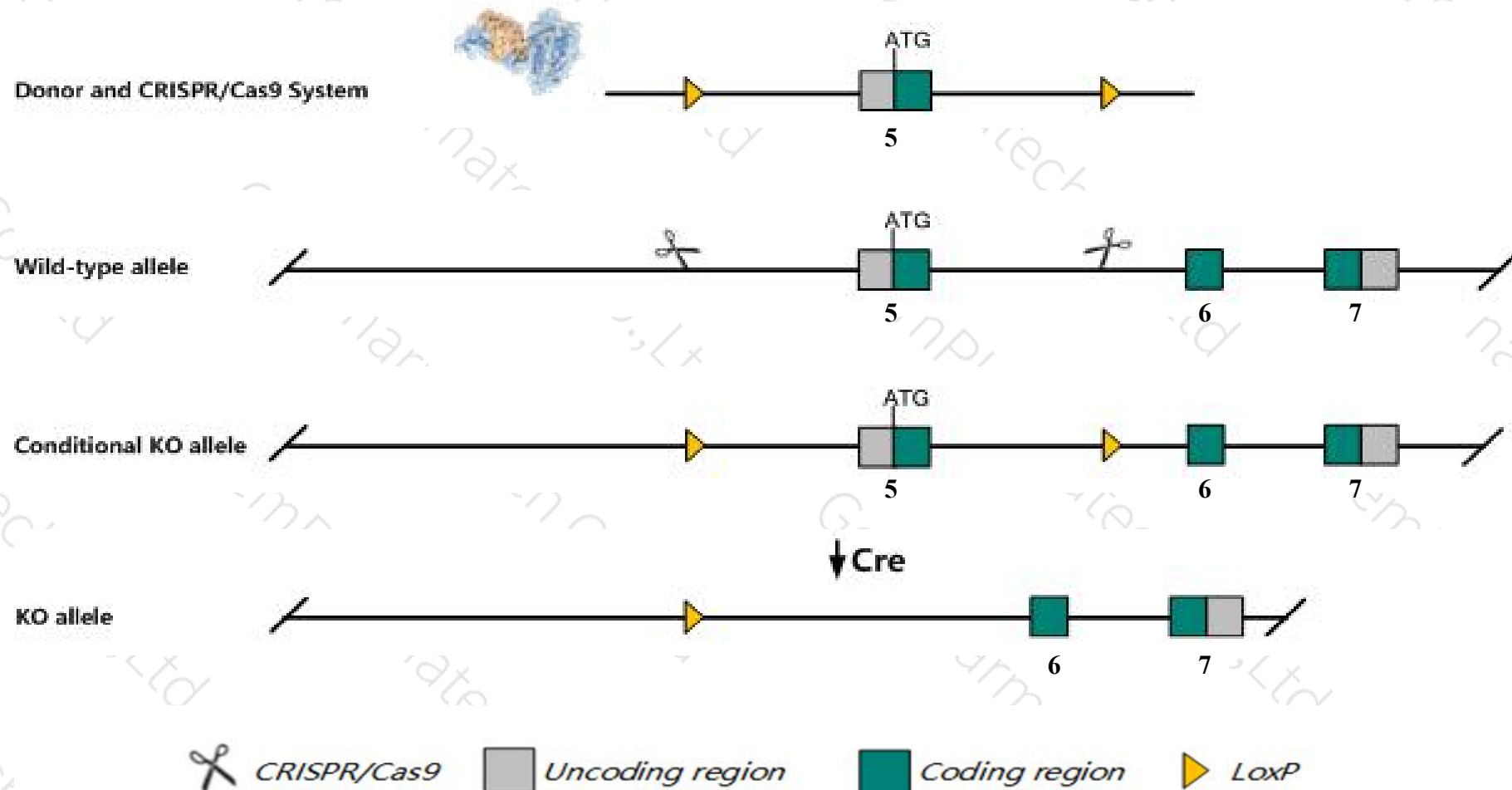|	
Project Name	*Ncald*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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

Technical routes

➢ The *Ncald* gene has 12 transcripts. According to the structure of *Ncald* gene, exon5 of *Ncald-201* (ENSMUST00000090150.10) transcript is recommended as the knockout region.The region contains start codon ATG. Knock out the region will result in disruption of protein function.

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

Notice

➢ According to the existing MGI data,Mice homozygous for a targeted allele could not be generated. Mice heterozygous for the targeted allele exhibit increased systemic arterial systolic blood pressure.

➢ The *Ncald* gene is located on the Chr15. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

Ncald neurocalcin delta [Mus musculus (house mouse)]
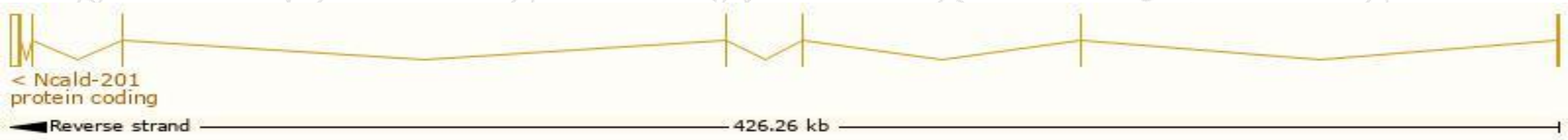
Gene ID: 52589, updated on 7-Apr-2019

▲Summary

Official Symbol	Ncald provided by MGI
Official Full Name	neurocalcin delta provided by MGI
Primary source	MGI:MGI:1196326
See related	Ensembl:ENSMUSG00000051359
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	AI848120, D030020D09Rik, D15Ertd412e
Expression	Biased expression in cortex adult (RPKM 51.2), frontal lobe adult (RPKM 39.6) and 11 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

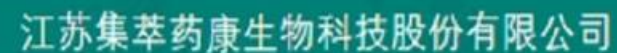The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ncald-201	ENSMUST00000090150.10	3733	193aa	Protein coding	CCDS27435	Q91X97	TSL:1 GENCODE basic APPRIS P1
Ncald-211	ENSMUST00000168992.7	3538	193aa	Protein coding	CCDS27435	Q91X97	TSL:5 GENCODE basic APPRIS P1
Ncald-204	ENSMUST00000120746.7	3407	193aa	Protein coding	CCDS27435	Q91X97	TSL:1 GENCODE basic APPRIS P1
Ncald-203	ENSMUST00000119730.7	3295	193aa	Protein coding	CCDS27435	Q91X97	TSL:1 GENCODE basic APPRIS P1
Ncald-202	ENSMUST00000116445.8	858	193aa	Protein coding	CCDS27435	Q91X97	TSL:3 GENCODE basic APPRIS P1
Ncald-210	ENSMUST00000153775.8	794	187aa	Protein coding	-	D3YVA2	CDS 3' incomplete TSL:5
Ncald-208	ENSMUST00000148652.8	787	161aa	Protein coding	-	D3Z2Z8	CDS 3' incomplete TSL:3
Ncald-209	ENSMUST00000150453.1	720	88aa	Protein coding	-	D3Z1M0	CDS 3' incomplete TSL:3
Ncald-212	ENSMUST00000226924.1	1992	No protein	Retained intron	-	-	
Ncald-207	ENSMUST00000137944.7	2277	No protein	lncRNA	-	-	TSL:1
Ncald-206	ENSMUST00000132423.1	709	No protein	lncRNA	-	-	TSL:3
Ncald-205	ENSMUST00000123317.1	662	No protein	lncRNA	-	-	TSL:3

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

< Ncald-201
protein coding

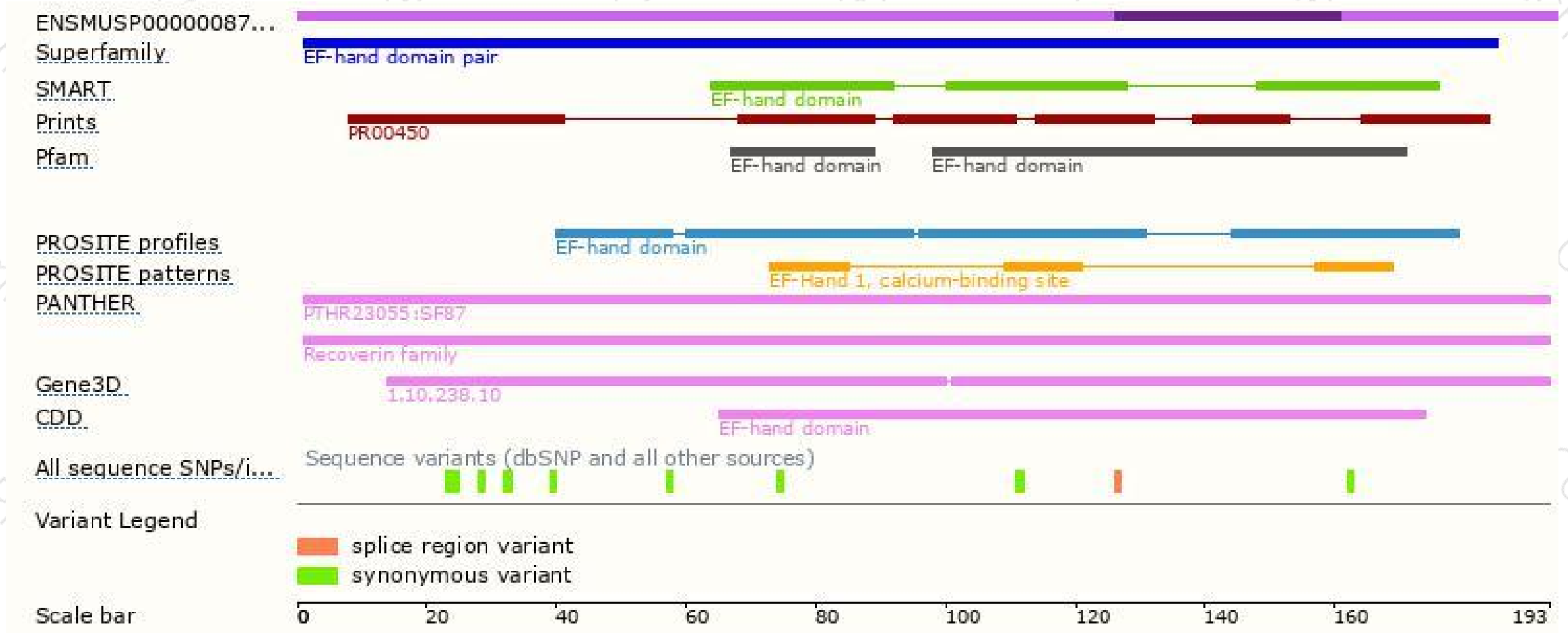◄— Reverse strand ———————————— 426.26 kb ————————————

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 targeted allele could not be generated. Mice heterozygous for the targeted allele exhibit increased systemic arterial systolic blood pressure.

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