

Ncam2 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Ncam2

Project type

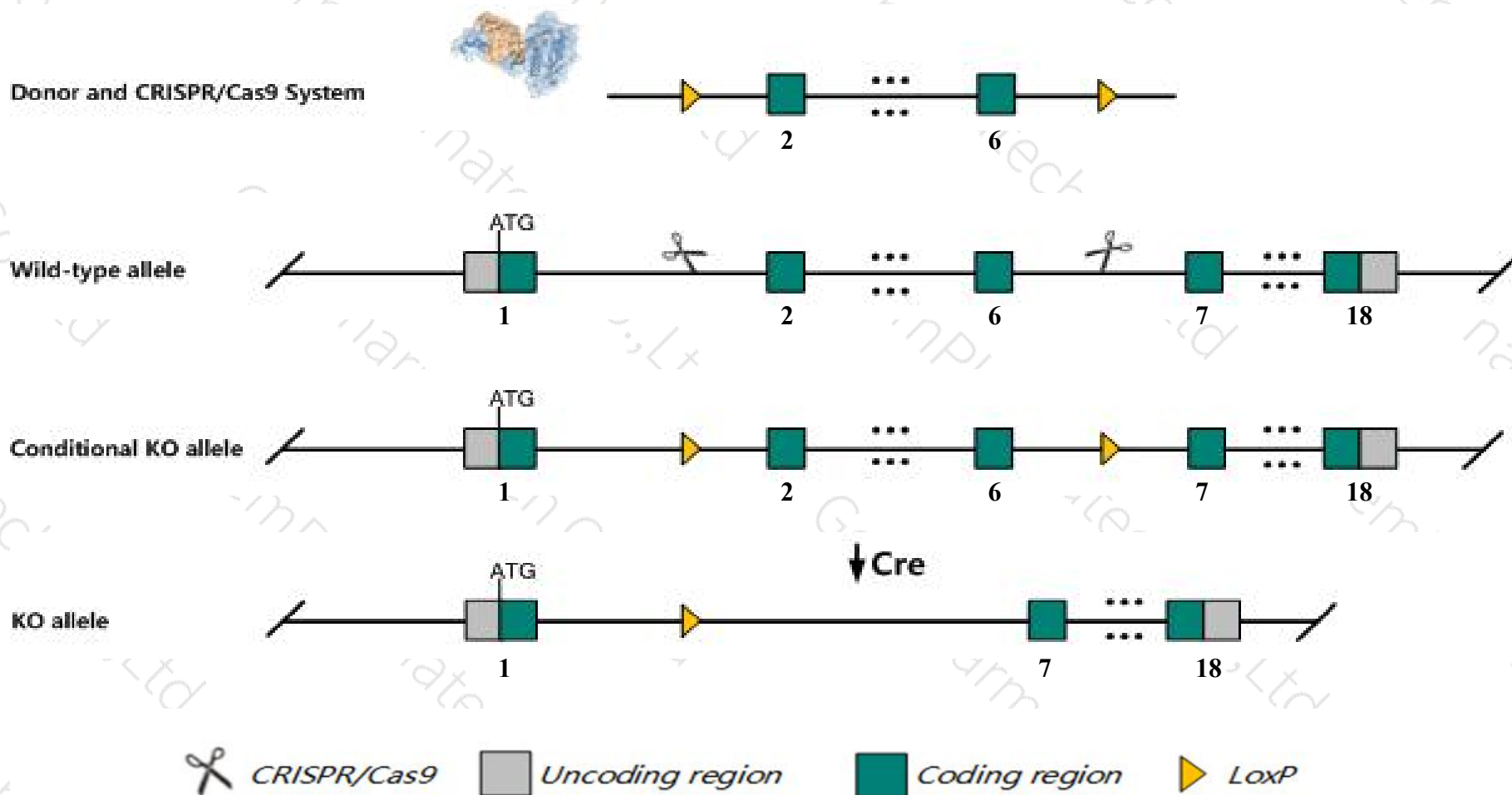
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ncam2* gene. The schematic diagram is as follows:



Technical routes

- The *Ncam2* gene has 4 transcripts. According to the structure of *Ncam2* gene, exon2-exon6 of *Ncam2*-202 (ENSMUST00000067602.4) transcript is recommended as the knockout region. The region contains 682bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ncam2* 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.

- According to the existing MGI data, A gene trap insertion into an intron of this gene results in no obvious phenotype. Mice homozygous for a knock-out allele exhibit increased proliferation rate and clonogenic frequency in spinal cord-derived neurospheres.
- The *Ncam2* gene is located on the Chr16. 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)

Ncam2 neural cell adhesion molecule 2 [Mus musculus (house mouse)]

Gene ID: 17968, updated on 31-Jan-2019

Summary



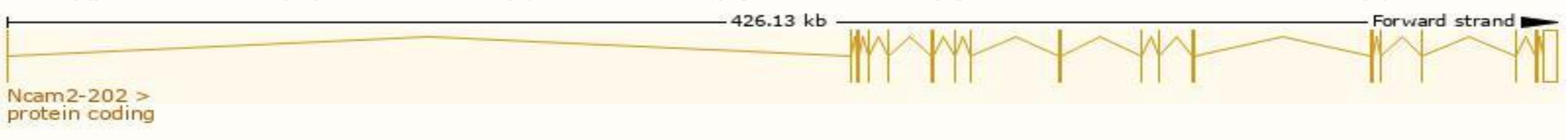
Official Symbol	Ncam2 provided by MGI
Official Full Name	neural cell adhesion molecule 2 provided by MGI
Primary source	MGI:MGI:97282
See related	Ensembl:ENSMUSG00000022762
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Ncam-2, Ocam, RNCAM
Expression	Biased expression in cortex adult (RPKM 3.2), frontal lobe adult (RPKM 3.0) and 6 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

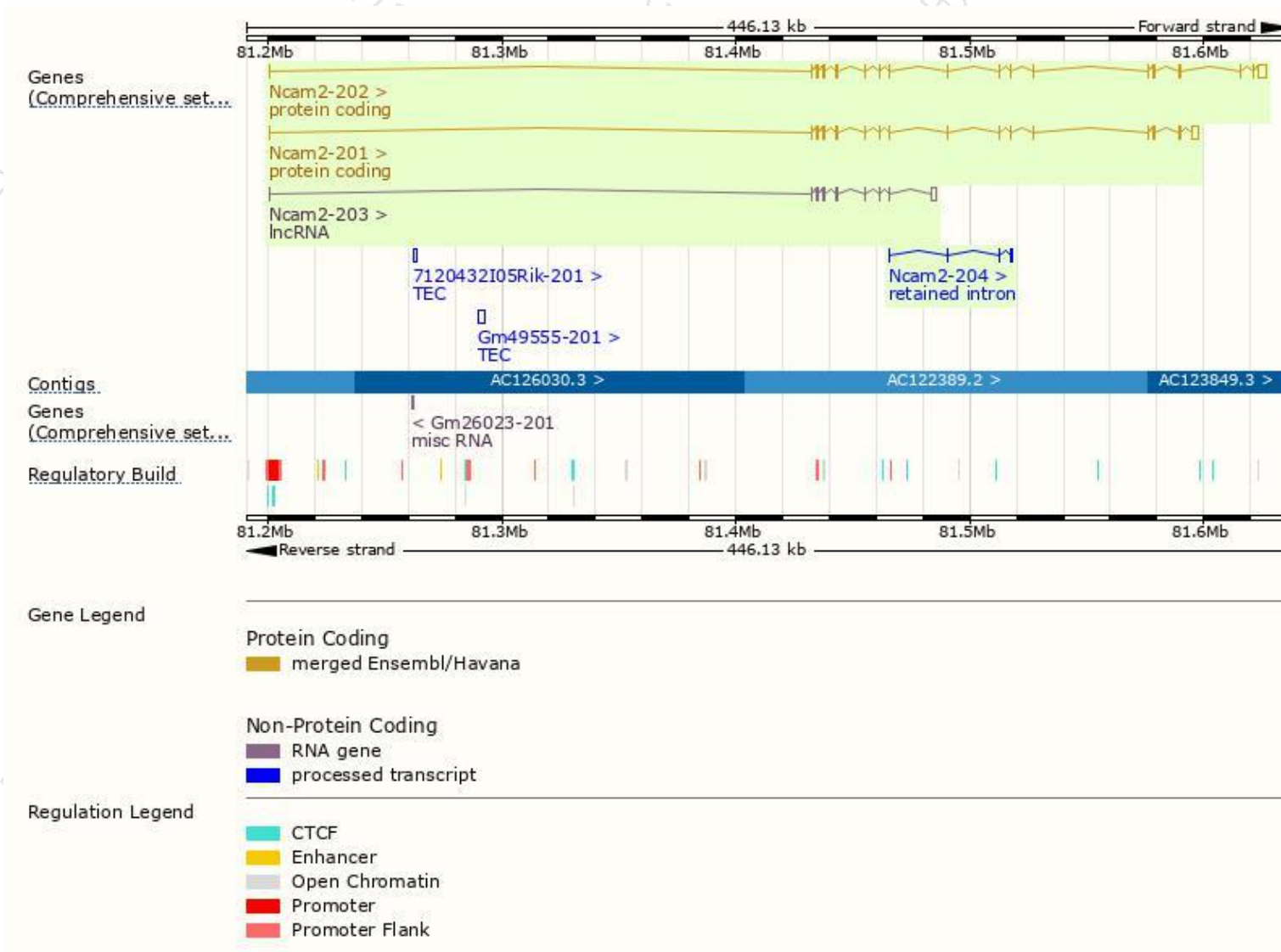
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ncam2-202	ENSMUST00000067602.4	6106	837aa	Protein coding	CCDS49888	Q35136	TSL:1 GENCODE basic APPRIS ALT2
Ncam2-201	ENSMUST00000037785.13	4893	727aa	Protein coding	CCDS28281	Q35136	TSL:1 GENCODE basic APPRIS P3
Ncam2-204	ENSMUST00000232550.1	931	No protein	Retained intron	-	-	
Ncam2-203	ENSMUST00000231687.1	3413	No protein	lncRNA	-	-	

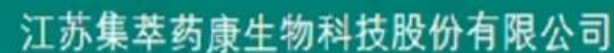
The strategy is based on the design of *Ncam2-202* transcript,The transcription is shown below



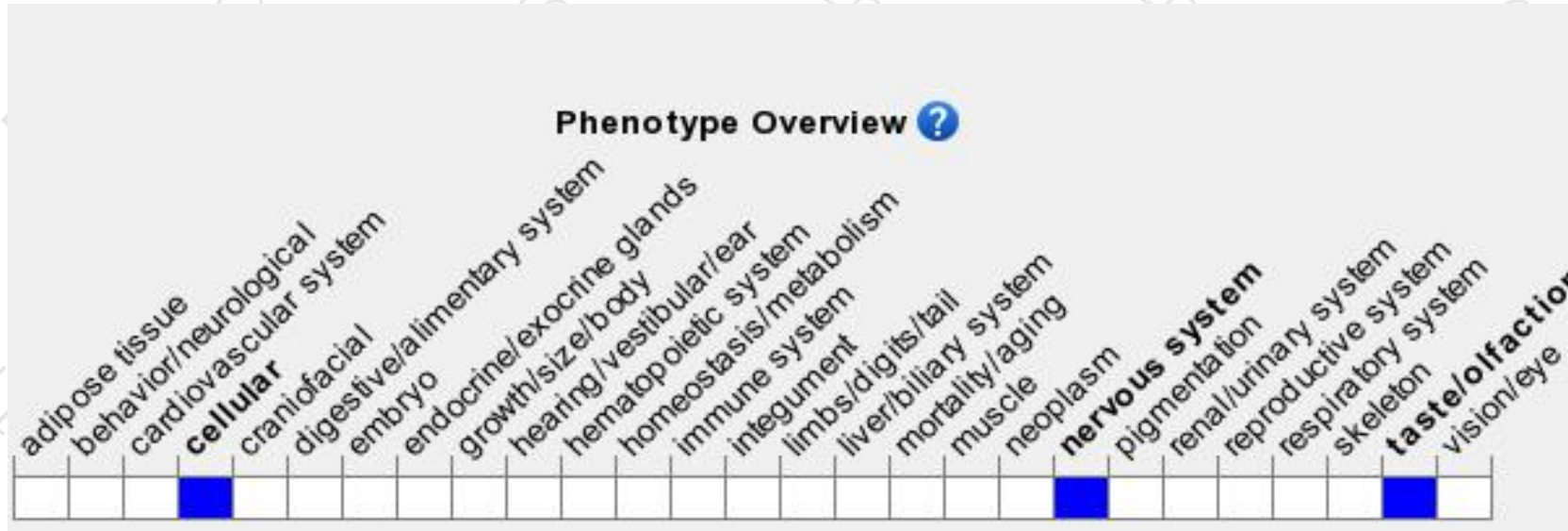
Genomic location distribution



集萃药康
GemPharmatech



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, A gene trap insertion into an intron of this gene results in no obvious phenotype.

Mice homozygous for a knock-out allele exhibit increased proliferation rate and clonogenic frequency in spinal cord-derived neurospheres.

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

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