

Nrcam Cas9-CKO Strategy

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Design Date: 2019-11-26

Project Overview



Project Name

Nrcam

Project type

Cas9-CKO

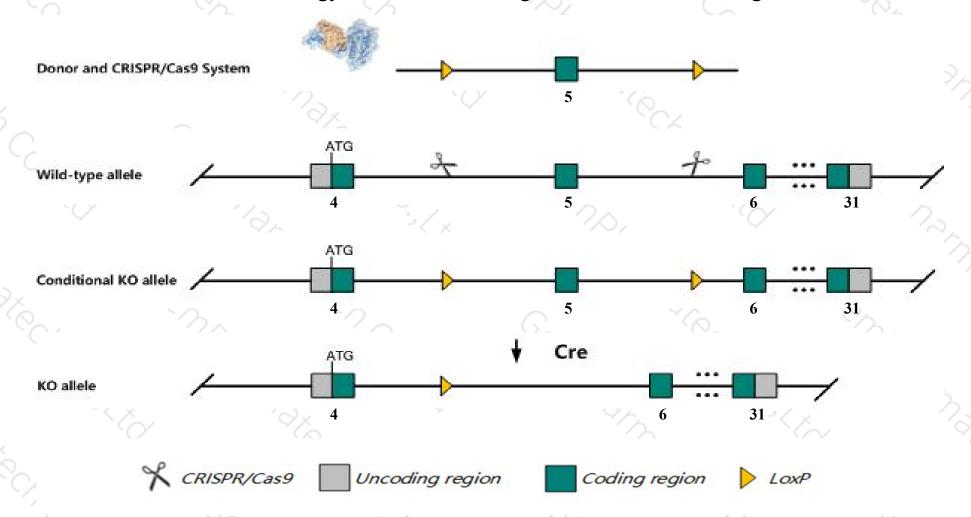
Strain background

C57BL/6JGpt

Conditional Knockout strategy



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



Technical routes



- ➤ The *Nrcam* gene has 17 transcripts. According to the structure of *Nrcam* gene, exon5 of *Nrcam-201*(ENSMUST00000020939.15) transcript is recommended as the knockout region. The region contains 106bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Nrcam* 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, Homozygotes for targeted null mutations exhibit disorganization of lens fibers, cellular disintegration, and accumulation of cellular debris resulting in cataracts. Mutants show mild reductions in cerebellar lobe size.
- ➤ Transcript Nrcam-205/214 CDS are incomplete, whether they will be affected is unknown.
- The *Nrcam* gene is located on the Chr12. 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)



Nrcam neuronal cell adhesion molecule [Mus musculus (house mouse)]

Gene ID: 319504, updated on 12-Aug-2019

Summary

2 ?

Official Symbol Nrcam provided by MGI

Official Full Name neuronal cell adhesion molecule provided by MGI

Primary source MGI:MGI:104750

See related Ensembl:ENSMUSG00000020598

Gene type protein coding
RefSeq status VALIDATED
Organism Mus musculus

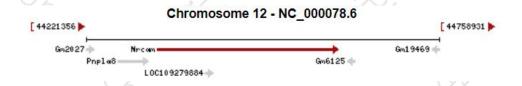
Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as Bravo; mKIAA0343; C030017F07Rik; C130076O07Rik

Expression Biased expression in cortex adult (RPKM 15.8), frontal lobe adult (RPKM 14.8) and 6 other tissues See more

Orthologs human all



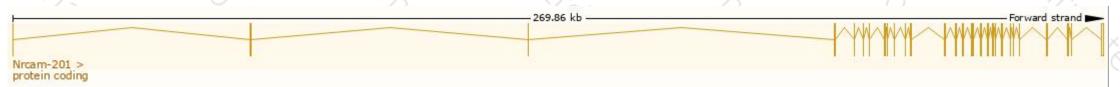
Transcript information (Ensembl)



The gene has 17 transcripts, all transcripts are shown below:

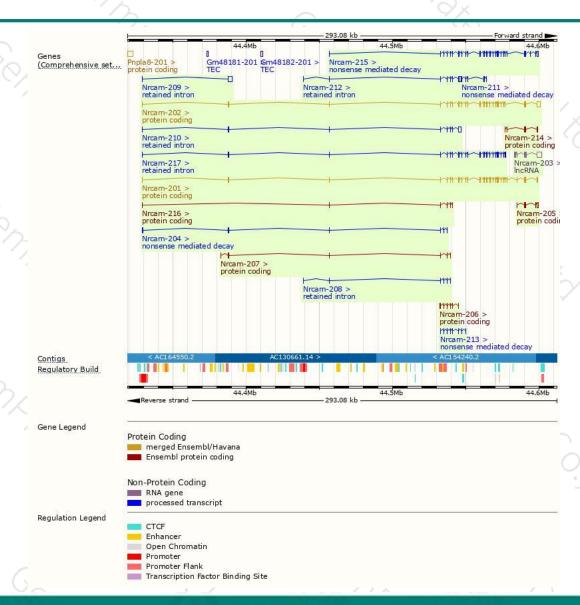
	··· /· /	-		~ /). +	~ / /)		The same of the sa	A service of the serv
Name 🌲	Transcript ID 🍦	bp 🍦	Protein 🌲	Translation ID 👙	Biotype	CCDS 🍦	UniProt 🍦	Flags
Nrcam-202	ENSMUST00000110748.3	6275	<u>1186aa</u>	ENSMUSP00000106376.2	Protein coding	CCDS49057₽	Q810U4₽	TSL:1 GENCODE basic APPRIS P1
Nrcam-201	ENSMUST00000020939.15	4469	1256aa	ENSMUSP00000020939.8	Protein coding	CCDS49056@	Q810U4®	TSL:1 GENCODE basic
Nrcam-205	ENSMUST00000218062.1	1586	168aa	ENSMUSP00000151475.1	Protein coding		<u>A0A1W2P6Z1</u> ₽	CDS 5' incomplete TSL:1
Nrcam-214	ENSMUST00000220082.1	916	220aa	ENSMUSP00000151824.1	Protein coding		A0A1W2P7X4日	CDS 5' incomplete TSL:1
Vrcam-206	ENSMUST00000218431.1	823	232aa	ENSMUSP00000151873.1	Protein coding		<u>A0A1W2P814</u> ₽	CDS 3' incomplete TSL:3
Ircam-216	ENSMUST00000220126.1	736	<u>175aa</u>	ENSMUSP00000151296.1	Protein coding		A0A1W2P6P3世	CDS 3' incomplete TSL:5
Ircam-207	ENSMUST00000218540.1	625	81aa	ENSMUSP00000151732.1	Protein coding		A0A1W2P7R6日	CDS 3' incomplete TSL:3
rcam-215	ENSMUST00000220123.1	4943	<u>1134aa</u>	ENSMUSP00000151844.1	Nonsense mediated decay		A0A1W2P7Y9世	TSL:1
Vrcam-213	ENSMUST00000219939.1	939	30aa	ENSMUSP00000152002.1	Nonsense mediated decay		A0A1W2P8F5₽	CDS 5' incomplete TSL:5
Ircam-204	ENSMUST00000217907.1	682	<u>59aa</u>	ENSMUSP00000151419.1	Nonsense mediated decay	5	A0A1W2P6V1&	CDS 5' incomplete TSL:5
Vrcam-211	ENSMUST00000219906.1	546	<u>41aa</u>	ENSMUSP00000151243.1	Nonsense mediated decay	5	A0A1W2P6F8₺	CDS 5' incomplete TSL:3
Ircam-217	ENSMUST00000220130.1	4176	No protein	-	Retained intron	-		TSL:1
Ircam-210	ENSMUST00000219592.1	3159	No protein	-	Retained intron	5	-0	TSL:5
Vrcam-209	ENSMUST00000218940.1	2610	No protein	-	Retained intron	5	-0	TSL:1
Ircam-212	ENSMUST00000219928.1	2433	No protein	-	Retained intron	5	-	TSL:2
Ircam-208	ENSMUST00000218805.1	677	No protein	-	Retained intron			TSL:3
Vrcam-203	ENSMUST00000217796.1	4487	No protein	-	IncRNA	5	70	TSL:5

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



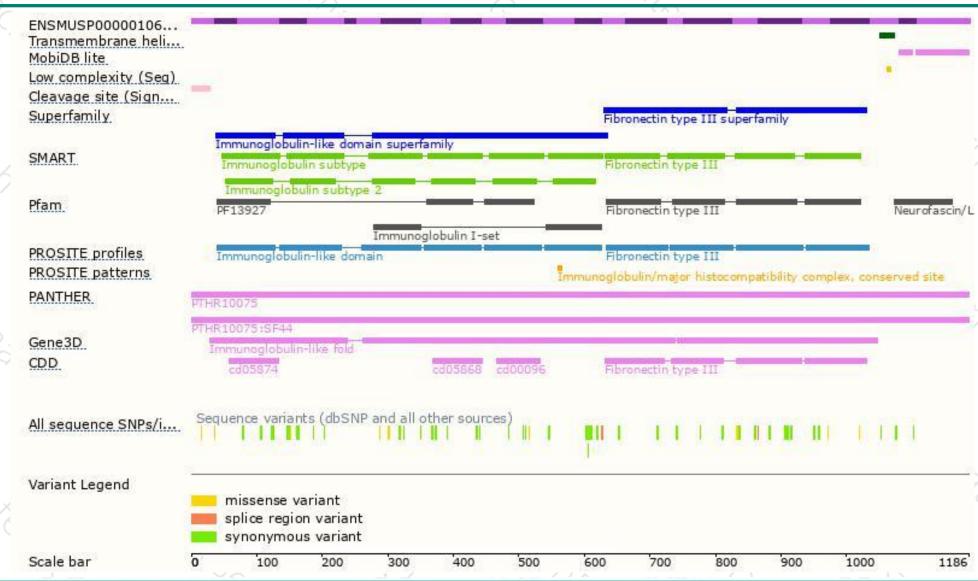
Genomic location distribution





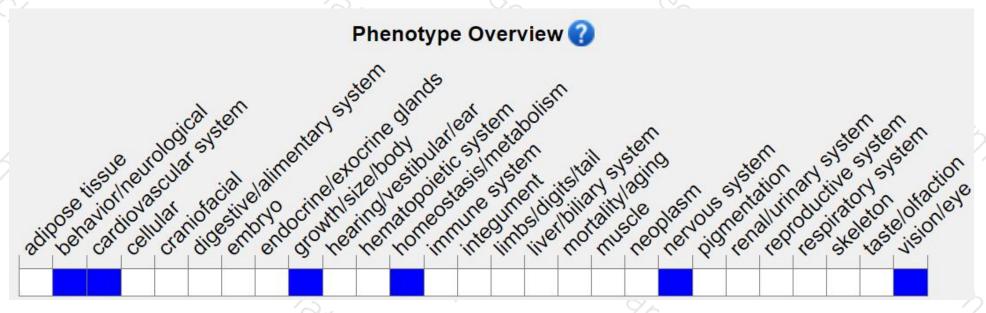
Protein domain





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, Homozygotes for targeted null mutations exhibit disorganization of lens fibers, cellular disintegration, and accumulation of cellular debris resulting in cataracts. Mutants show mild reductions in cerebellar lobe size.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





