

***Grik2* Cas9-CKO Strategy**

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Date: 2019-11-18

Project Overview

Project Name

Grik2

Project type

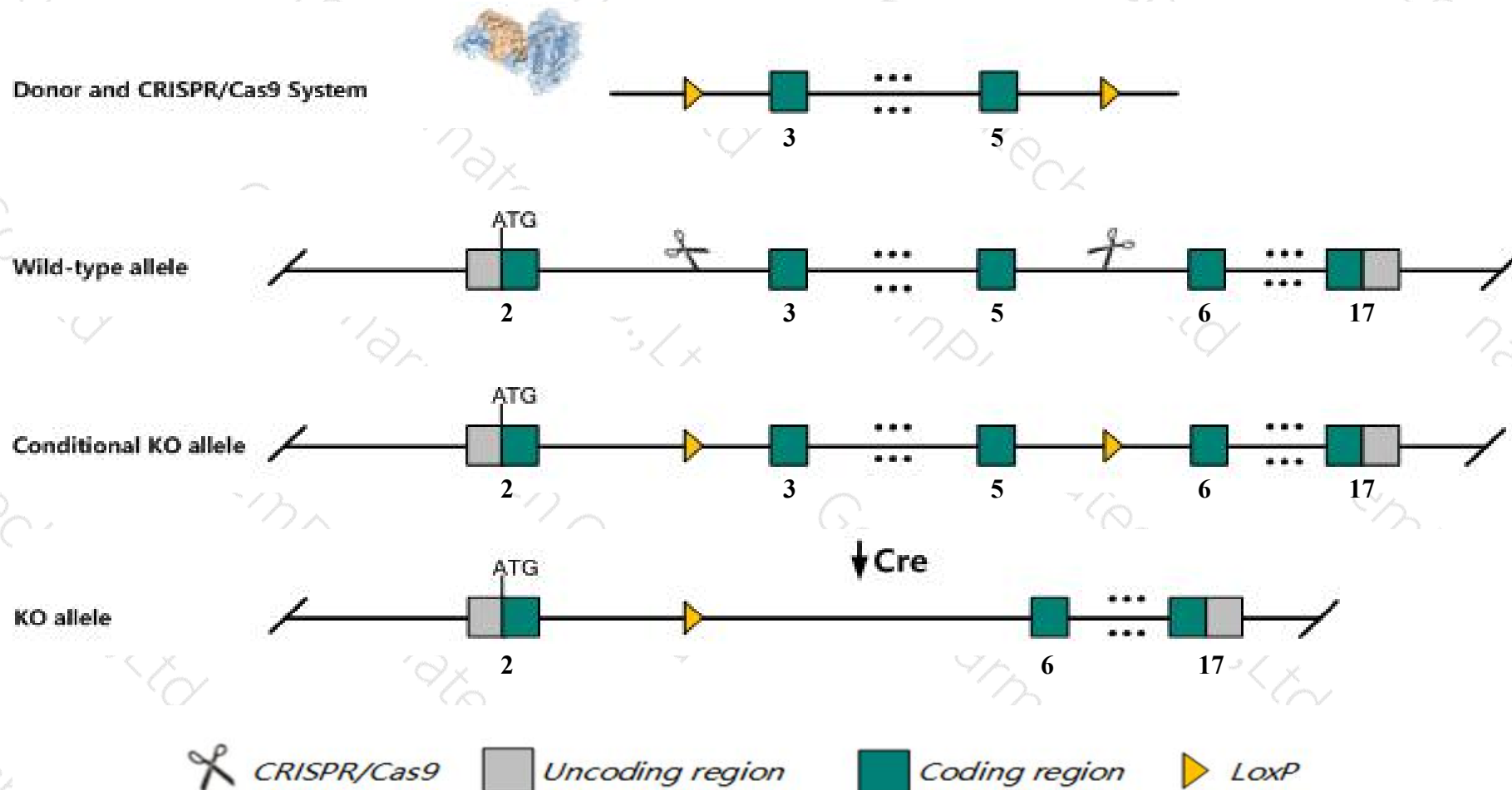
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Grik2* gene. The schematic diagram is as follows:



Technical routes

- The *Grik2* gene has 11 transcripts. According to the structure of *Grik2* gene, exon3-exon5 of *Grik2*-207 (ENSMUST00000218823.1) transcript is recommended as the knockout region. The region contains 608bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Grik2* 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 and cell types.

- According to the existing MGI data, Homozygotes for a targeted null mutation exhibit hippocampal neurons with reduced sensitivity to kainate and reduced susceptibility to the seizure-inducing effects of kainate administration.
- Transcript *Grik2*-203&208&211 may not be affected.
- The effect on transcript *Grik2*-206&209 is unknown.
- The *Grik2* gene is located on the Chr10. 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)

Grik2 glutamate receptor, ionotropic, kainate 2 (beta 2) [*Mus musculus* (house mouse)]

Gene ID: 14806, updated on 24-Oct-2019

Summary

Official Symbol	Grik2 provided by MGI
Official Full Name	glutamate receptor, ionotropic, kainate 2 (beta 2) provided by MGI
Primary source	MGI:MGI:95815
See related	Ensembl:ENSMUSG00000056073
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Gluk2; Glur6; Glur-6; AW124492; Glurbeta2; C130030K03Rik
Summary	Glutamate receptors are the predominant excitatory neurotransmitter receptors in the mammalian brain and are activated in a variety of normal neurophysiologic processes. This gene product belongs to the kainate family of glutamate receptors, which are composed of four subunits and function as ligand-activated ion channels. The subunit encoded by this gene is subject to RNA editing at multiple sites within the first and second transmembrane domains, which is thought to alter the structure and function of the receptor complex. Alternatively spliced transcript variants encoding different isoforms have also been found for this gene. [provided by RefSeq, Jul 2008]
Expression	Biased expression in CNS E18 (RPKM 9.9), whole brain E14.5 (RPKM 6.6) and 5 other tissues See more
Orthologs	human all

Genomic context

Location: 10 B3; 10 24.87 cM

Exon count: 23

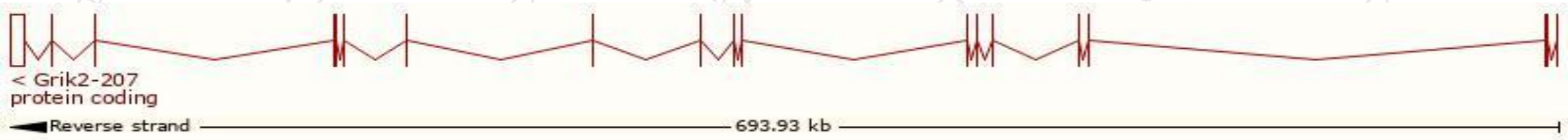
See Grik2 in [Genome Data Viewer](#)

Transcript information (Ensembl)

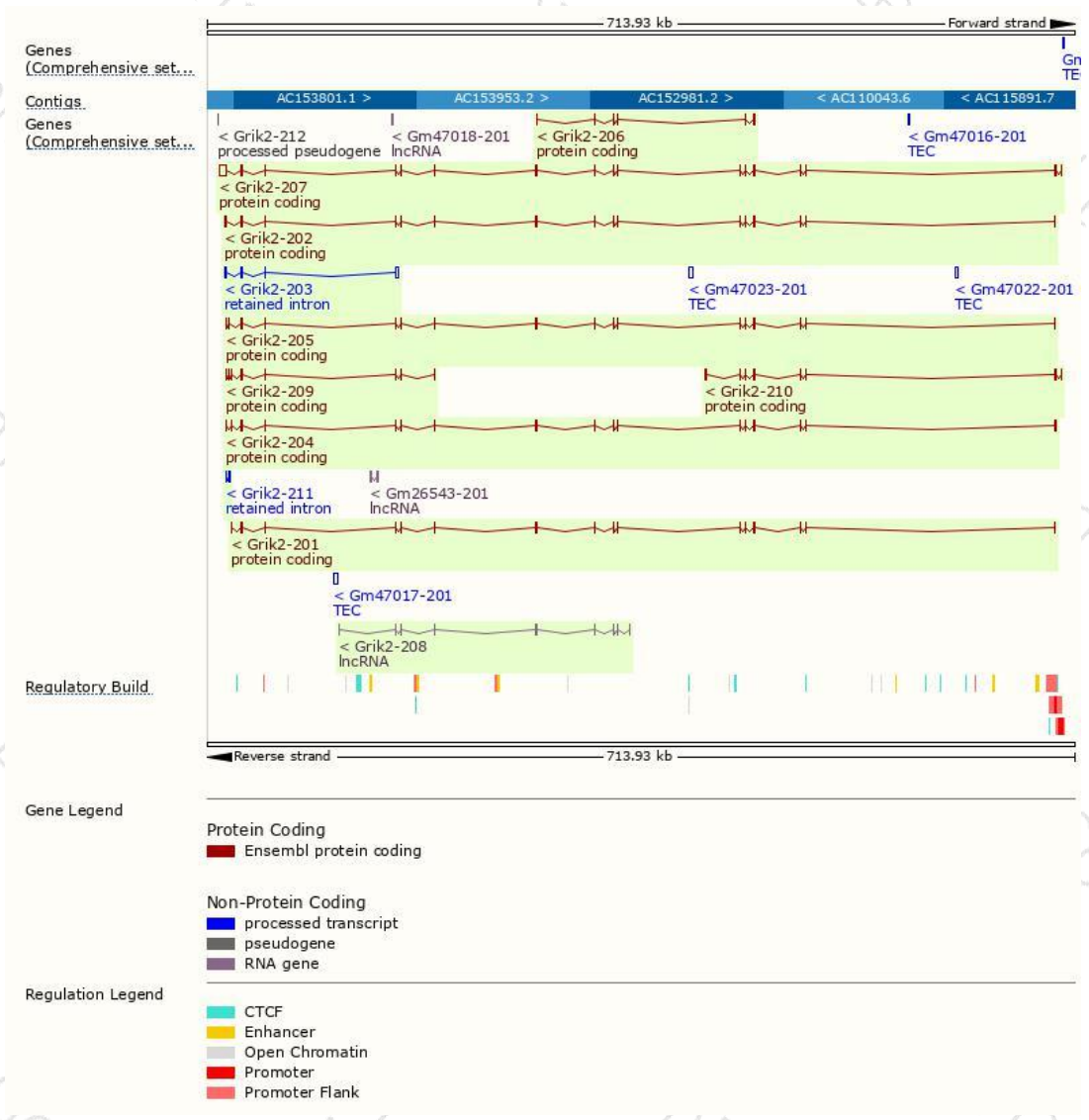
The gene has 11 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Grik2-207	ENSMUST00000218823.1	9427	908aa	Protein coding	CCDS48554	P39087	TSL:1 GENCODE basic APPRIS P1
Grik2-202	ENSMUST00000105484.9	3992	908aa	Protein coding	CCDS48554	P39087	TSL:1 GENCODE basic APPRIS P1
Grik2-204	ENSMUST00000218441.1	3083	869aa	Protein coding	CCDS23830	P39087	TSL:1 GENCODE basic
Grik2-201	ENSMUST00000079751.8	2610	869aa	Protein coding	CCDS23830	P39087	TSL:2 GENCODE basic
Grik2-205	ENSMUST00000218598.1	3245	893aa	Protein coding	-	A0A1W2P6S5	TSL:5 GENCODE basic
Grik2-210	ENSMUST00000220263.1	2085	338aa	Protein coding	-	Q6PAQ0	TSL:1 GENCODE basic
Grik2-209	ENSMUST00000219509.1	1542	309aa	Protein coding	-	A0A1W2P868	CDS 5' incomplete TSL:5
Grik2-206	ENSMUST00000218669.1	539	179aa	Protein coding	-	A0A1W2P8D9	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Grik2-203	ENSMUST00000217673.1	4365	No protein	Retained intron	-	-	TSL:1
Grik2-211	ENSMUST00000220330.1	567	No protein	Retained intron	-	-	TSL:3
Grik2-208	ENSMUST00000219051.1	1374	No protein	lncRNA	-	-	TSL:5

The strategy is based on the design of *Grik2-207* transcript,The transcription is shown below



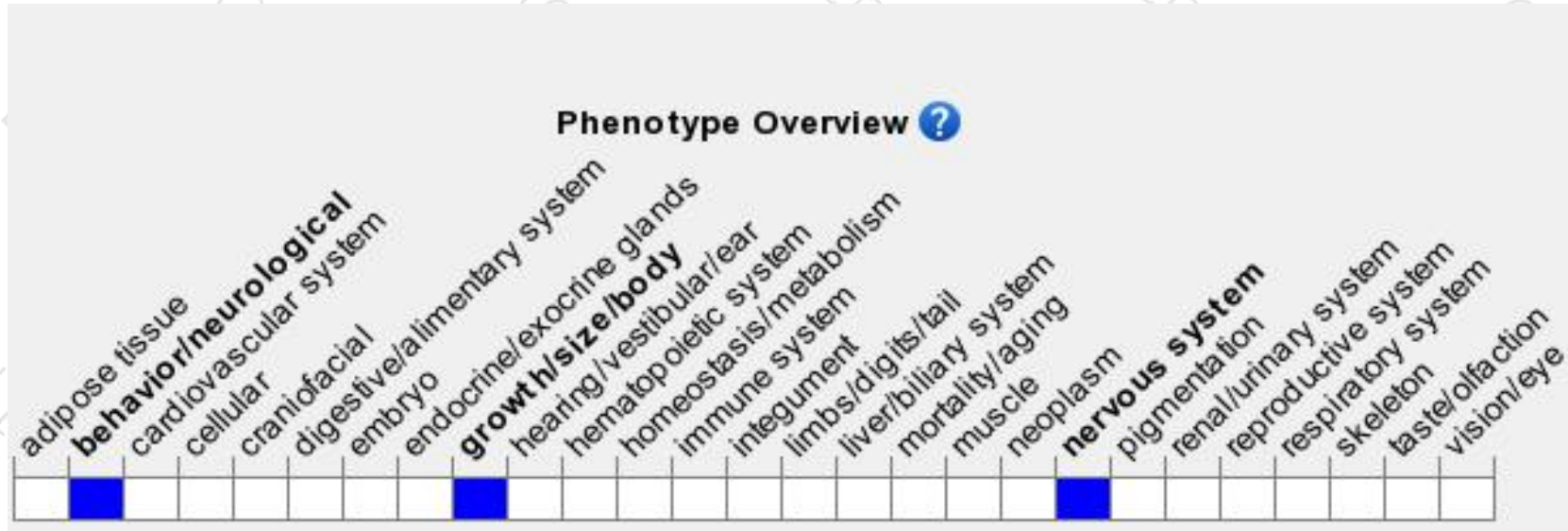
Genomic location distribution



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 a targeted null mutation exhibit hippocampal neurons with reduced sensitivity to kainate and reduced susceptibility to the seizure-inducing effects of kainate administration.

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

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