Grin2b Cas9-CKO Strategy

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Design Date: 2019-7-18

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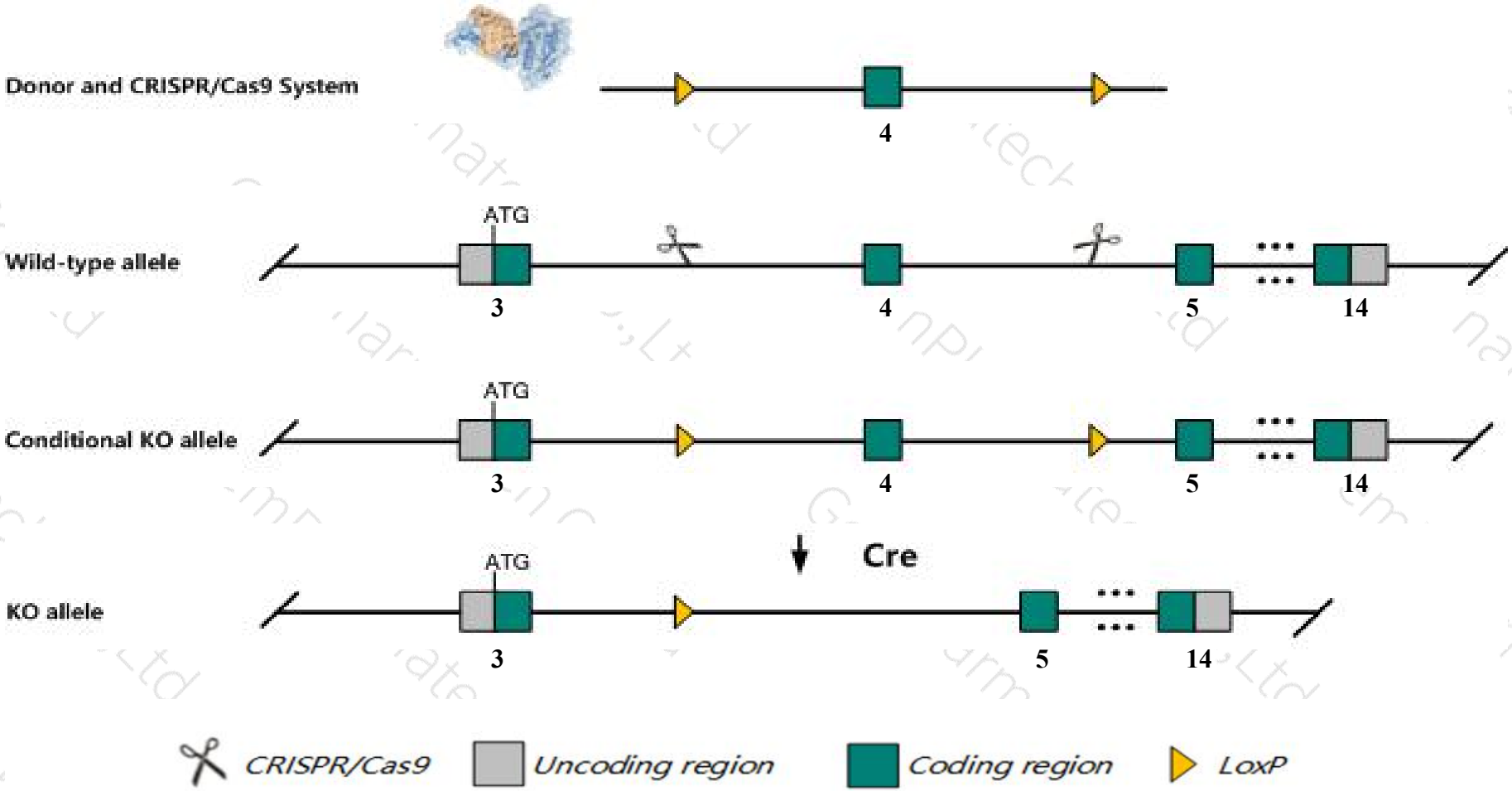
Project Overview

Project Name	***Grin2b***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Grin2b* gene. The schematic diagram is as follows:



Technical routes

- The *Grin2b* gene has 7 transcripts. According to the structure of *Grin2b* gene, exon4 of *Grin2b-201* (ENSMUST00000053880.12) transcript is recommended as the knockout region.The region contains 599bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Grin2b* 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 impairments in suckling, in hippocampal long term depression, and in pattern formation of trigeminal nucleus sensory afferent terminals. Mutants die shortly after birth.

➢ The *Grin2b* gene is located on the Chr6. 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）

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Grin2b glutamate receptor, ionotropic, NMDA2B (epsilon 2) [Mus musculus (house mouse)]

Gene ID: 14812, updated on 9-Apr-2019

Summary

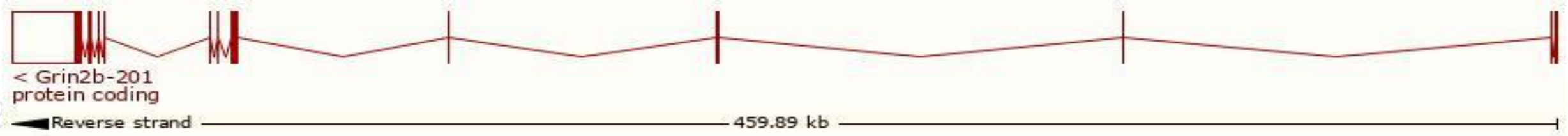
Official Symbol	Grin2b provided by MGI
Official Full Name	glutamate receptor, ionotropic, NMDA2B (epsilon 2) provided by MGI
Primary source	MGI:MGI:95821
See related	Ensembl:ENSMUSG00000030209
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AW490526, GluN2B, GluRepsilon2, NR2B, Nmdar2b
Expression	Biased expression in cortex adult (RPKM 5.7), frontal lobe adult (RPKM 5.7) and 5 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

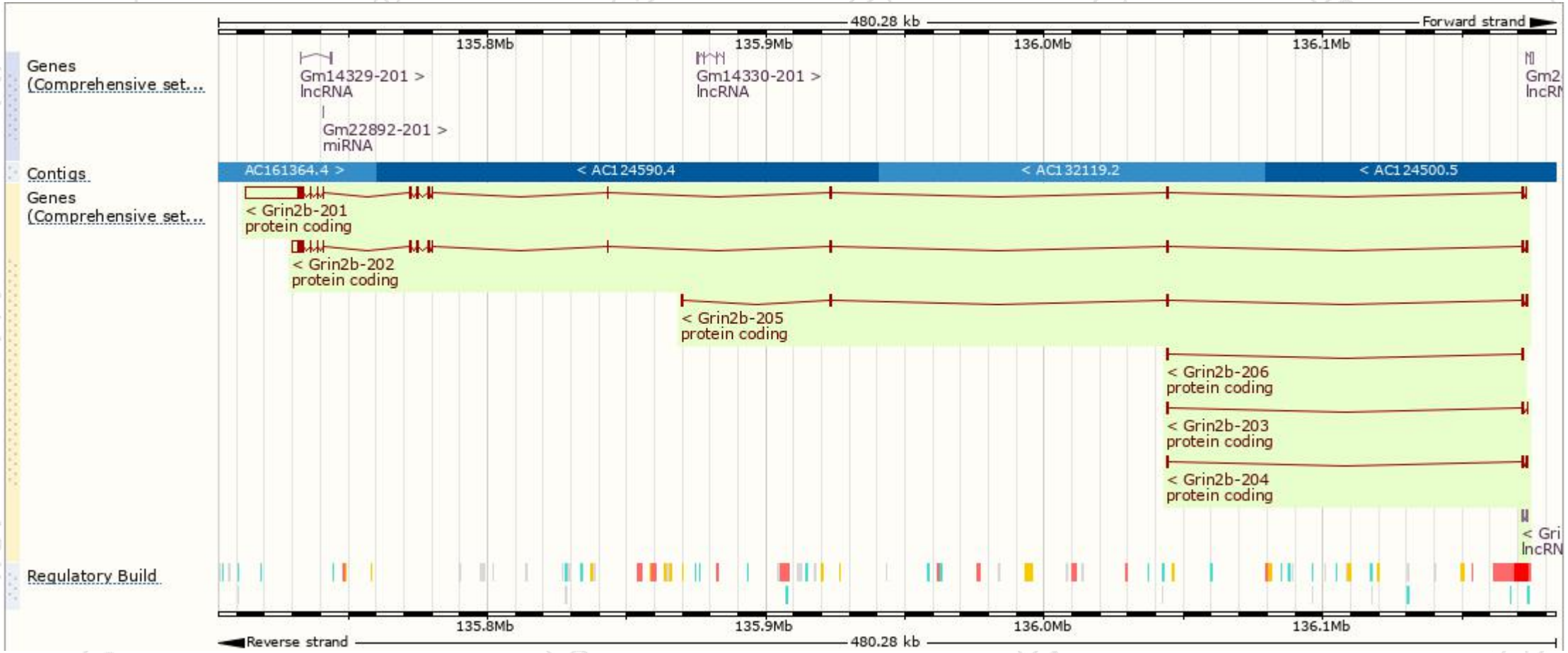
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Grin2b-201	ENSMUST00000053880.12	24060	1482aa	Protein coding	CCDS20648	G3X9V4	TSL:5 GENCODE basic APPRIS P1
Grin2b-202	ENSMUST00000111905.7	7484	1482aa	Protein coding	CCDS20648	G3X9V4	TSL:5 GENCODE basic APPRIS P1
Grin2b-205	ENSMUST00000152012.7	2461	337aa	Protein coding	-	A0A0G2JEA7	TSL:1 GENCODE basic
Grin2b-204	ENSMUST00000143943.7	703	35aa	Protein coding	-	Q8CG69	CDS 3' incomplete TSL:1
Grin2b-203	ENSMUST00000125905.1	630	35aa	Protein coding	-	Q8CG69	CDS 3' incomplete TSL:1
Grin2b-206	ENSMUST00000188999.2	601	36aa	Protein coding	-	A0A087WR33	CDS 3' incomplete TSL:1
Grin2b-207	ENSMUST00000198283.1	241	No protein	lncRNA	-	-	TSL:5

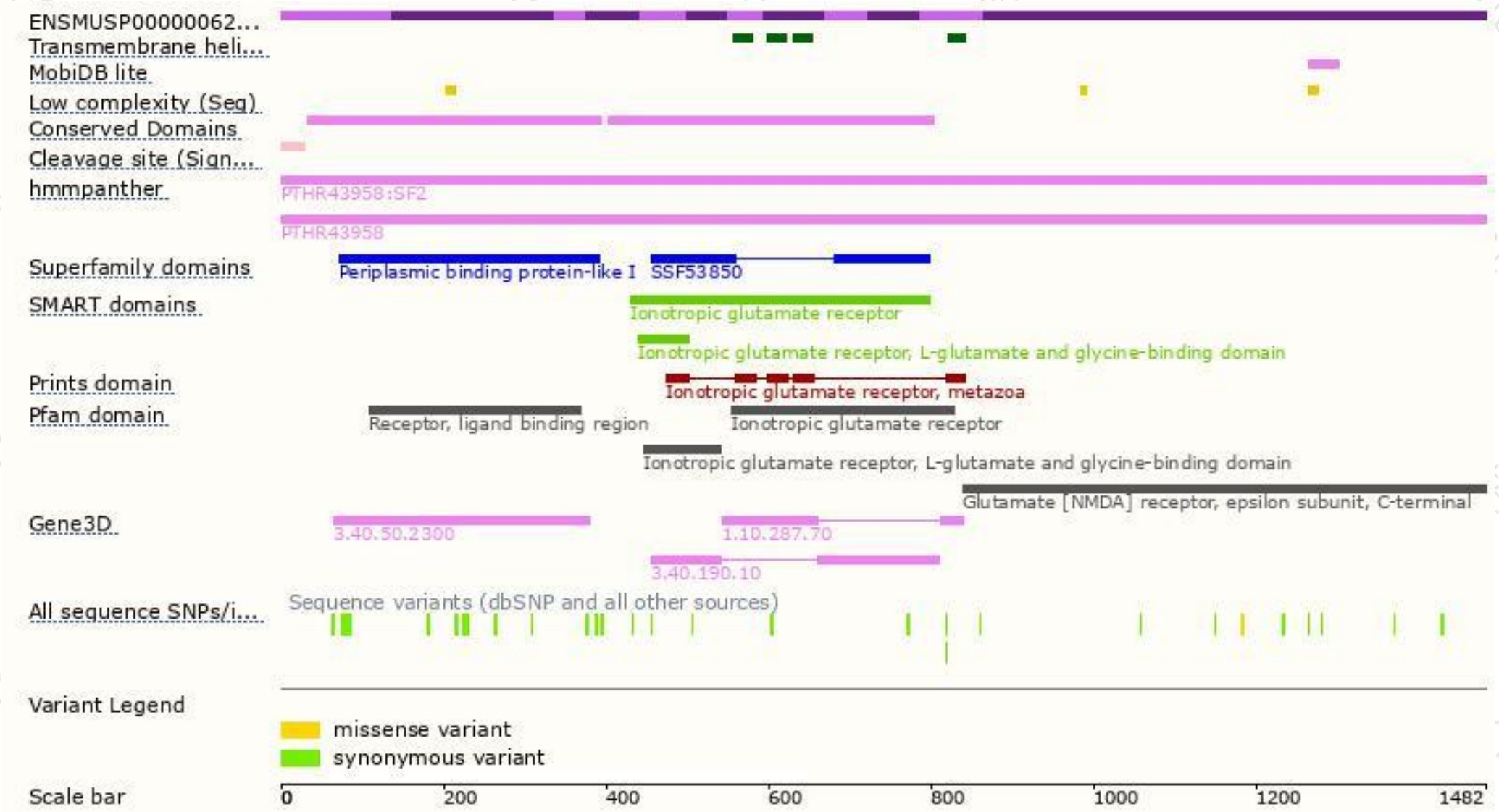
The strategy is based on the design of *Grin2b-201* transcript,The transcription is shown below



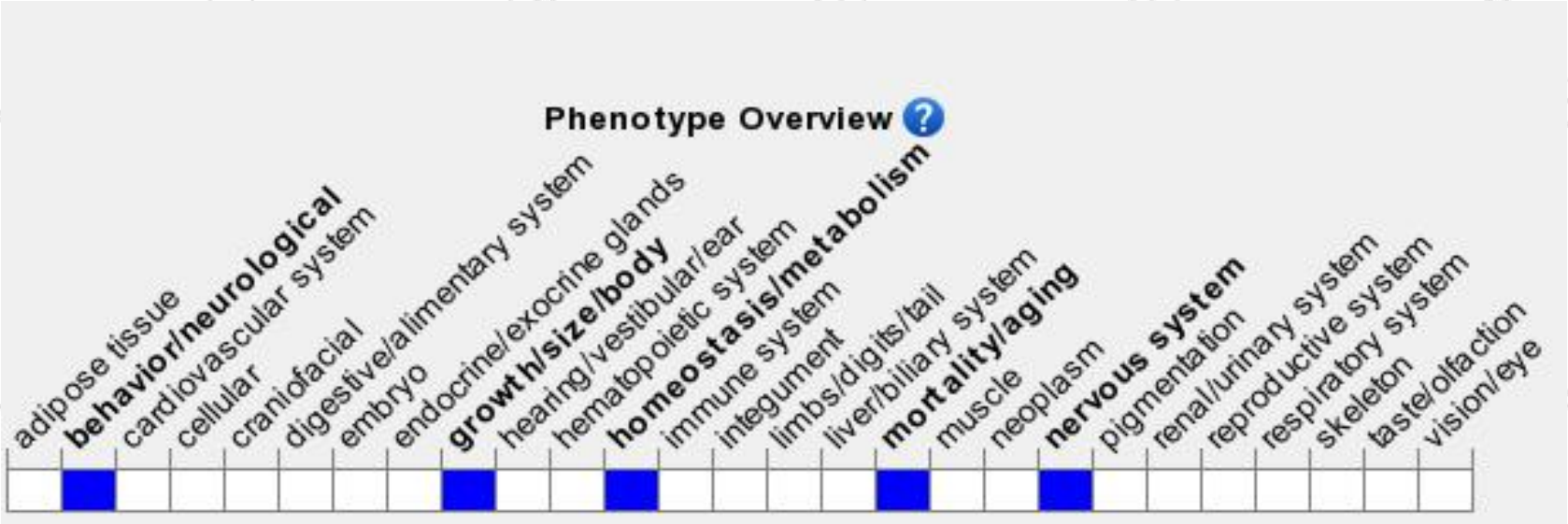
Genomic location distribution



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/).

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

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