

Hcn3 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Hcn3

Project type

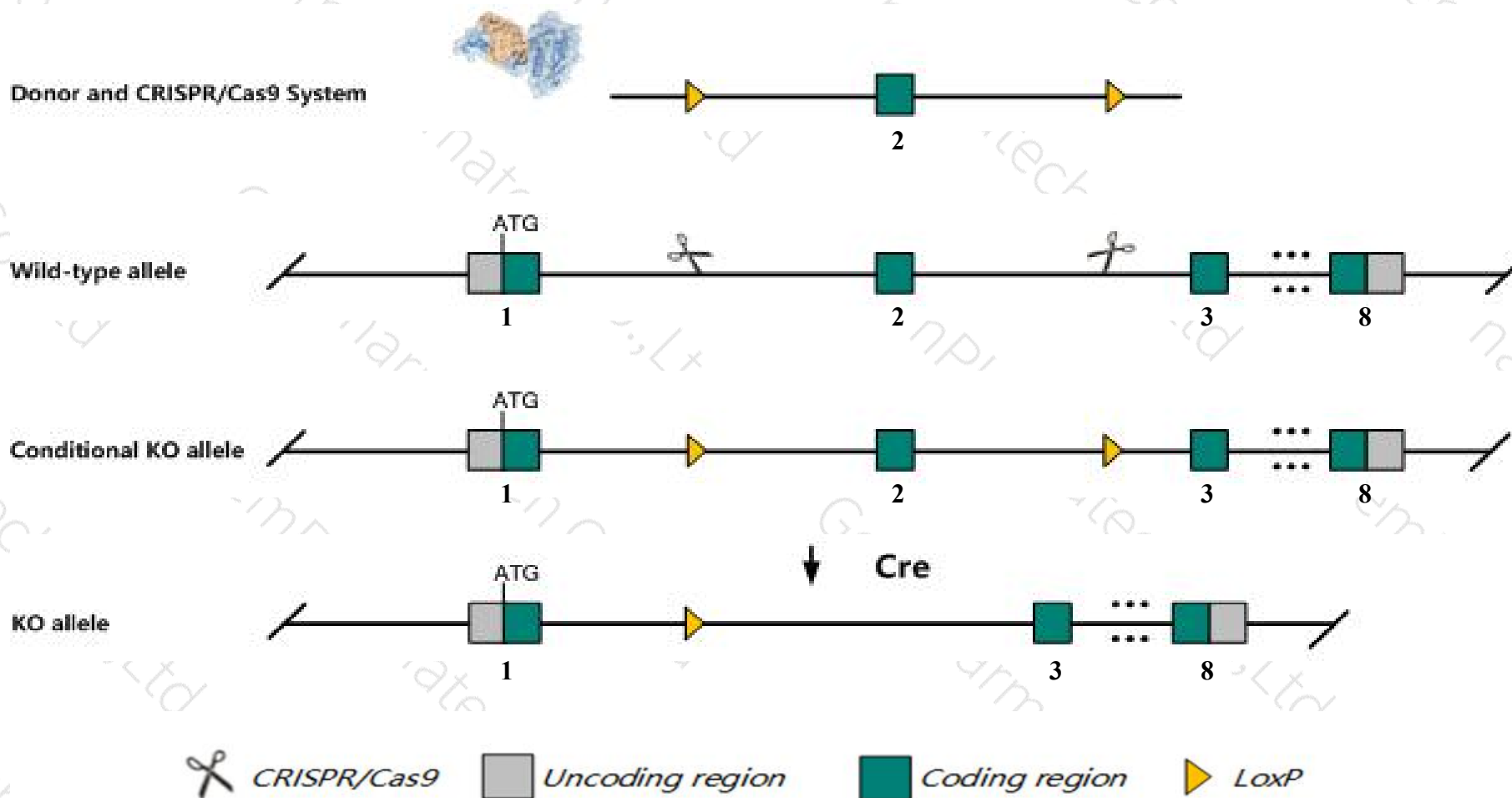
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hcn3* gene. The schematic diagram is as follows:



- The *Hcn3* gene has 4 transcripts. According to the structure of *Hcn3* gene, exon2 of *Hcn3-201* (ENSMUST00000029686.3) transcript is recommended as the knockout region. The region contains 430bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Hcn3* 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, Mice homozygous for a knock-out allele exhibit abnormal ventricular action potential waveform.
- The *Hcn3* gene is located on the Chr3. 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)

Hcn3 hyperpolarization-activated, cyclic nucleotide-gated K⁺ 3 [*Mus musculus* (house mouse)]

Gene ID: 15168, updated on 12-Aug-2019

Summary

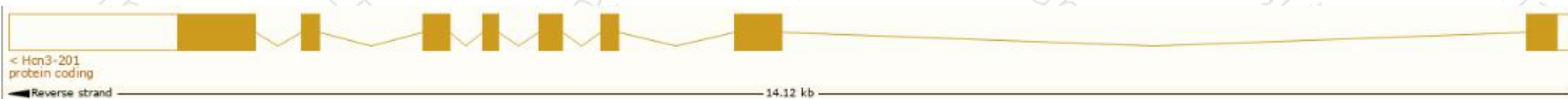
Official Symbol	Hcn3 provided by MGI
Official Full Name	hyperpolarization-activated, cyclic nucleotide-gated K ⁺ 3 provided by MGI
Primary source	MGI:MGI:1298211
See related	Ensembl:ENSMUSG00000028051
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Hac3; Hcn4; Bcng4; BCNG-4
Expression	Broad expression in CNS E18 (RPKM 16.2), whole brain E14.5 (RPKM 13.6) and 15 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

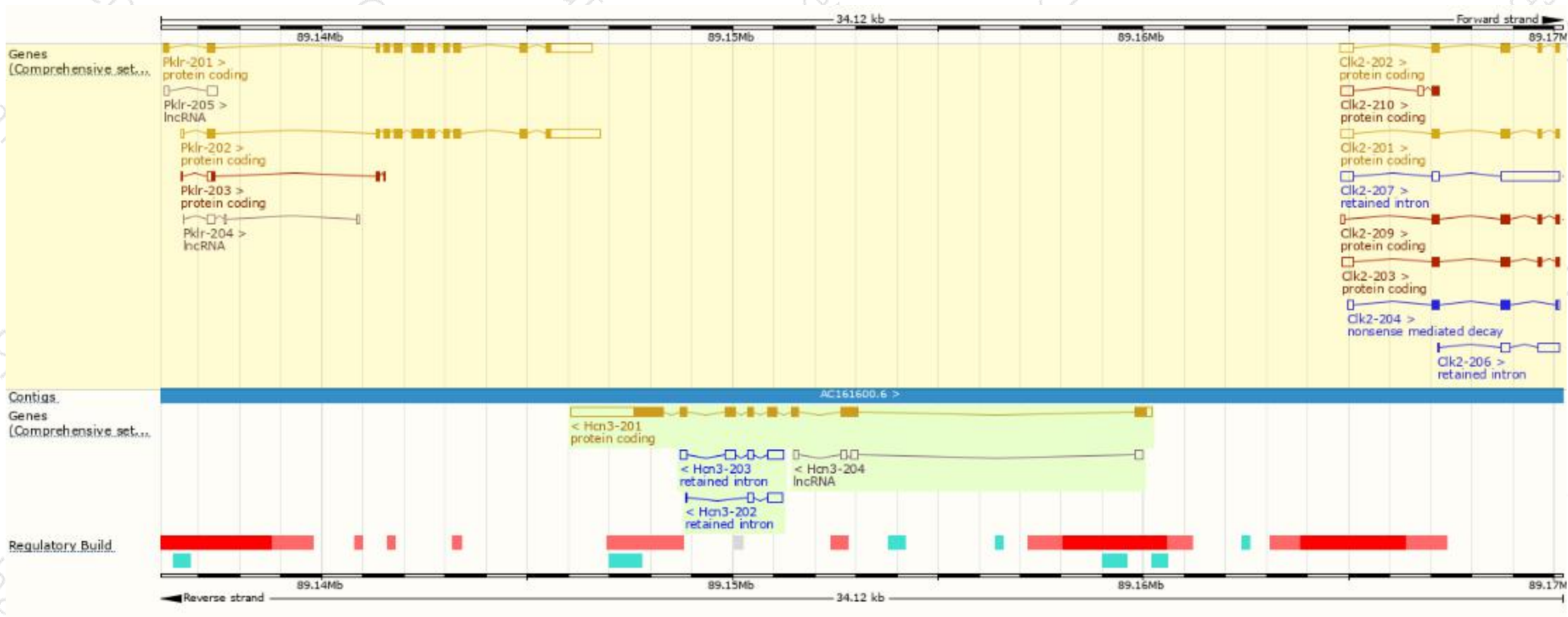
The gene has 4 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hcn3-201	ENSMUST00000029686.3	4002	779aa	Protein coding	CCDS17490	B2RRB5 O88705	TSL:1 GENCODE basic APPRIS P1
Hcn3-203	ENSMUST00000132156.7	909	No protein	Retained intron	-	-	TSL:5
Hcn3-202	ENSMUST00000127654.1	533	No protein	Retained intron	-	-	TSL:5
Hcn3-204	ENSMUST00000133368.1	627	No protein	lncRNA	-	-	TSL:3

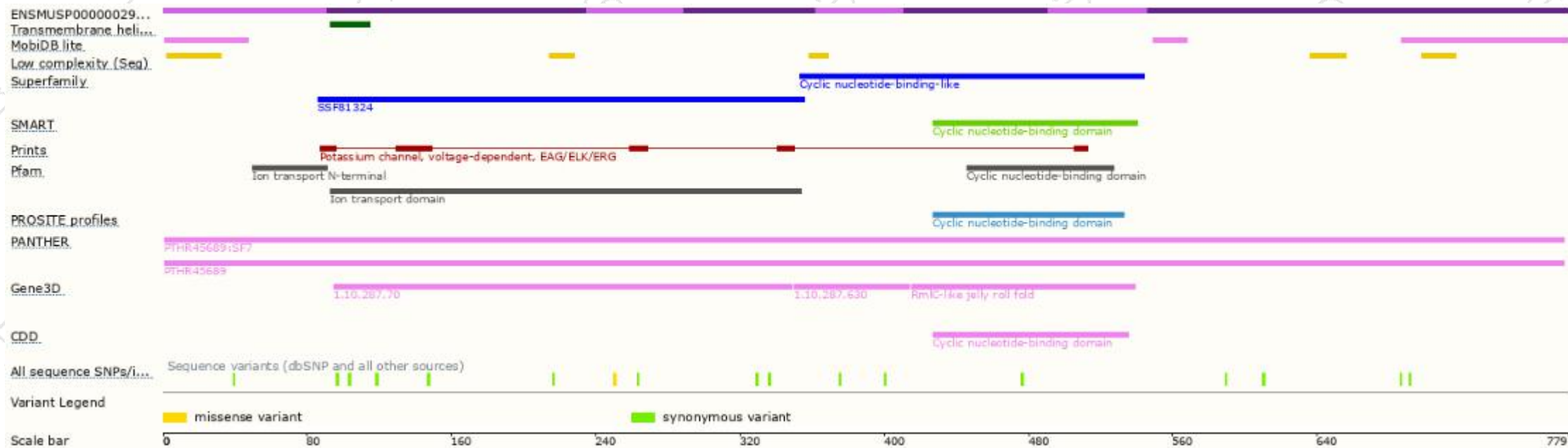
The strategy is based on the design of *Hcn3-201* transcript, The transcription is shown below



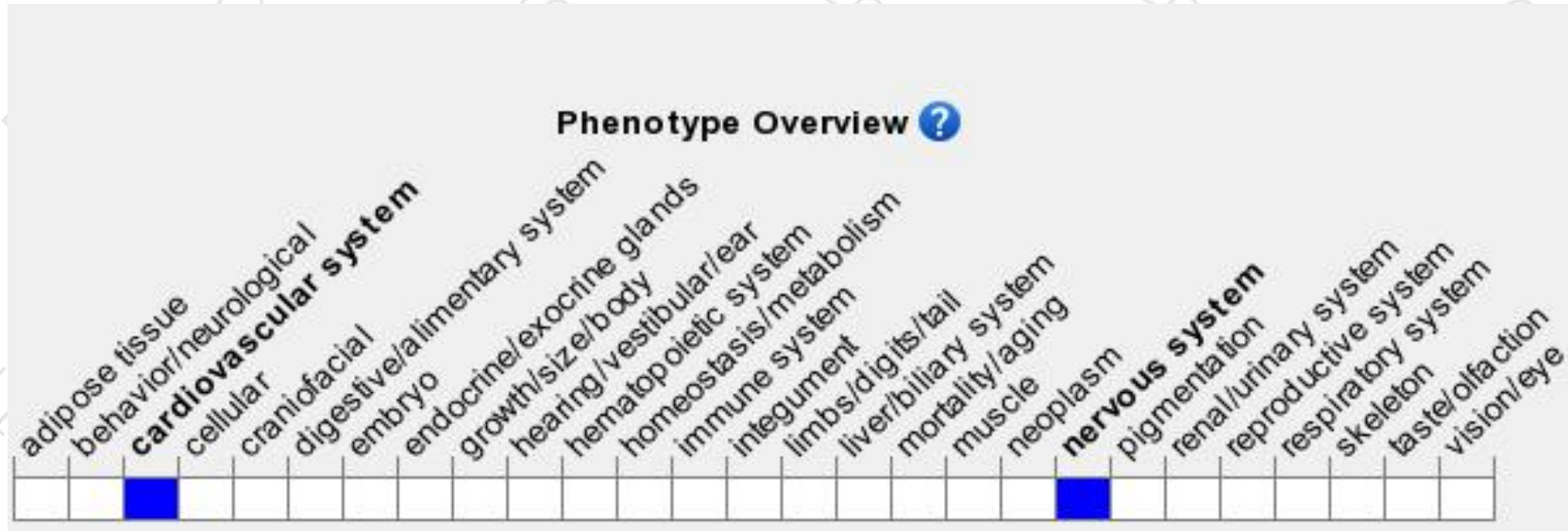
Genomic location distribution



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, Mice homozygous for a knock-out allele exhibit abnormal ventricular action potential waveform.

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

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