

Gpr4 Cas9-KO Strategy

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Reviewer:

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Design Date:

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Project Overview

Project Name

Gpr4

Project type

Cas9-KO

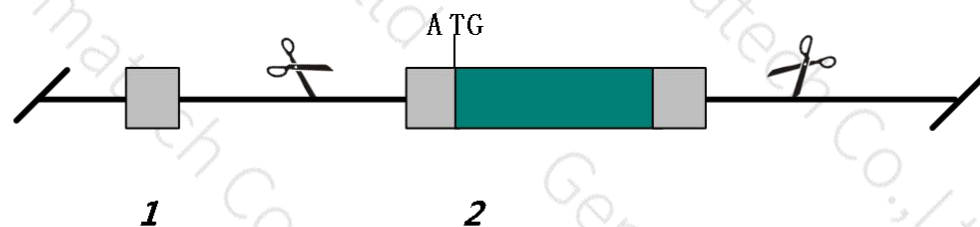
Strain background

C57BL/6JGpt

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Gpr4* gene. The schematic diagram is as follows:

Wild-type allele



KO allele



Cas9/sgRNA



Uncoding region



Coding region

Technical routes

- The *Gpr4* gene has 1 transcripts. According to the structure of *Gpr4* gene, exon2 of *Gpr4*-201 (ENSMUST00000060225.5)transcript is recommended as the knockout region.The region contains 1098bp all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Gpr4* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.

- According to the existing MGI data , mice homozygous for a null allele display partial neonatal and postnatal lethality, hemorrhages, impaired association of vascular smooth muscle cells with capillaries and small arteries and veins, and impaired contact between mesangial cells and renal glomerular capillaries.
- The xxx gene is located on the Chr7. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Gpr4 G protein-coupled receptor 4 [*Mus musculus* (house mouse)]

Gene ID: 319197, updated on 8-Dec-2018

Summary

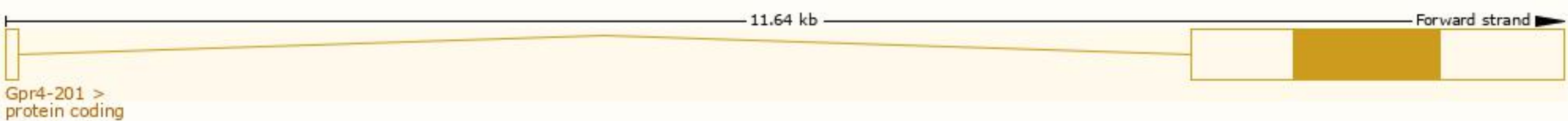
Official Symbol	Gpr4 provided by MGI
Official Full Name	G protein-coupled receptor 4 provided by MGI
Primary source	MGI:MGI:2441992
See related	Ensembl:ENSMUSG00000044317
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Ubiquitous expression in lung adult (RPKM 11.4), subcutaneous fat pad adult (RPKM 11.1) and 25 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

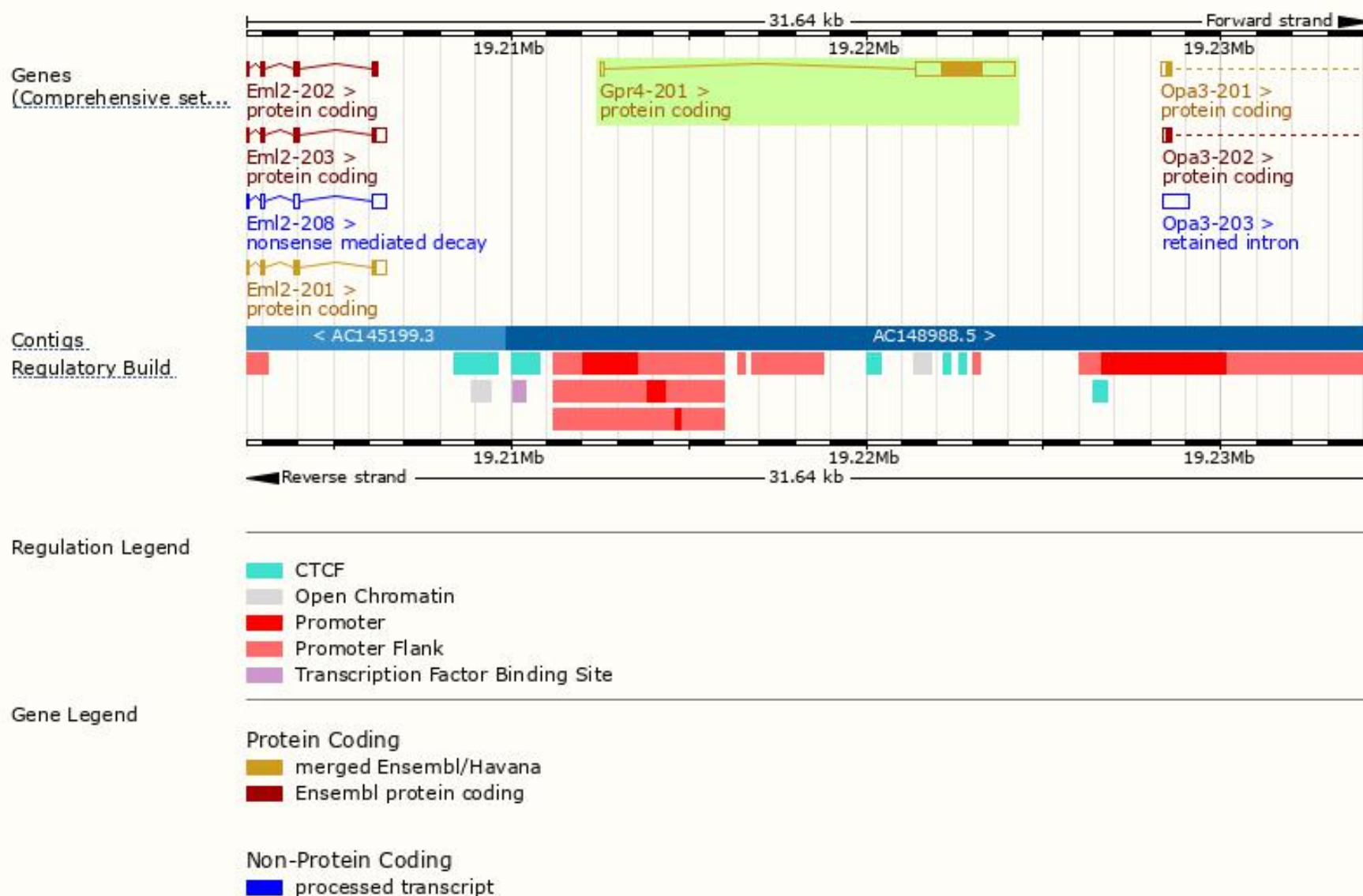
The gene has 1 transcript, and all transcripts are shown below:

Show/hide columns (1 hidden)								Filter	
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags	
Gpr4-201	ENSMUST00000060225.5	2882	365aa	Protein coding	CCDS20893	Q8BUD0	NM_175668 NP_783599	TSL:1	GENCODE basic APPRIS P1

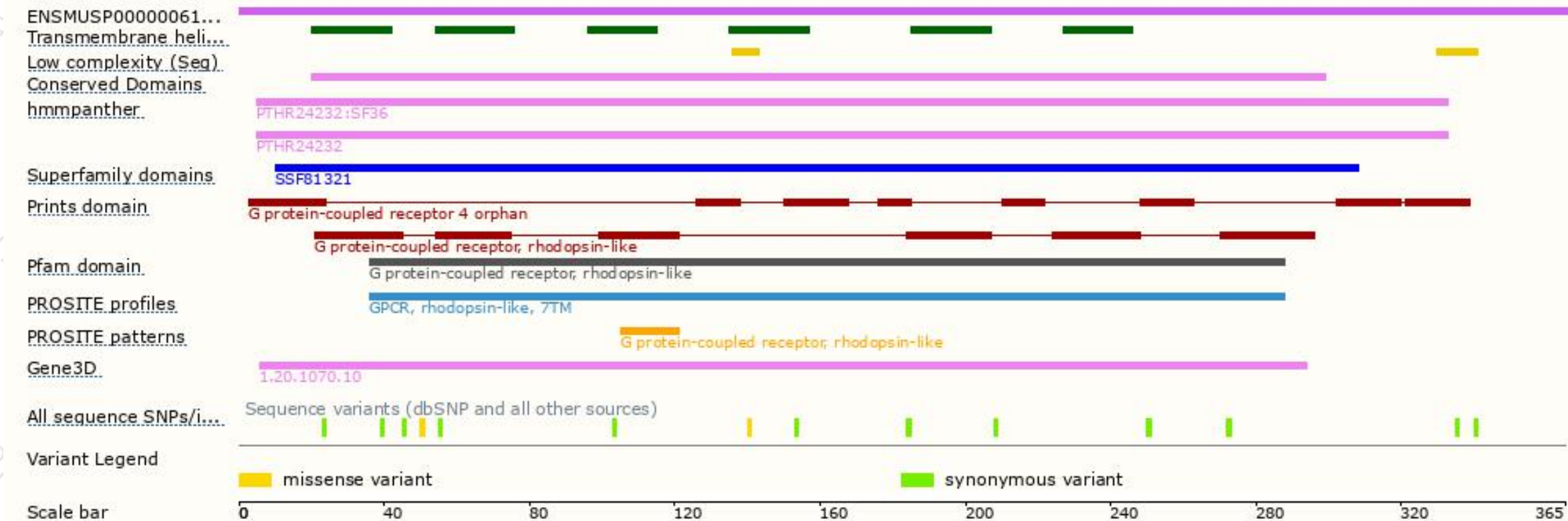
The strategy is based on the design of *Gpr4-201* transcript, The transcription is shown below



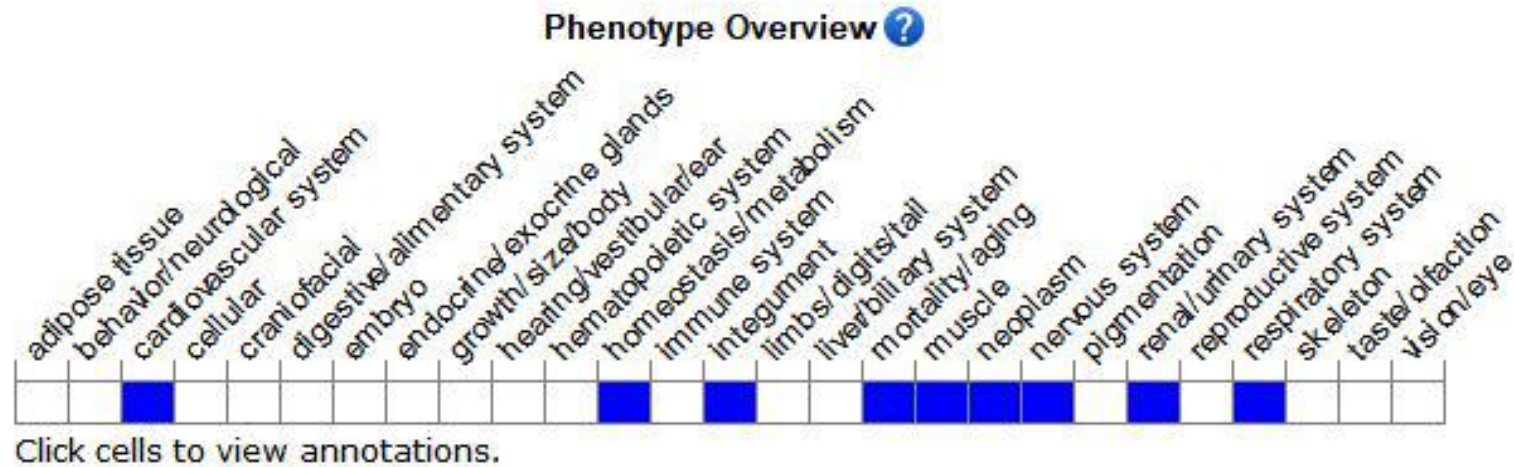
Genomic location distribution



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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