Htr2a Cas9-CKO Strategy

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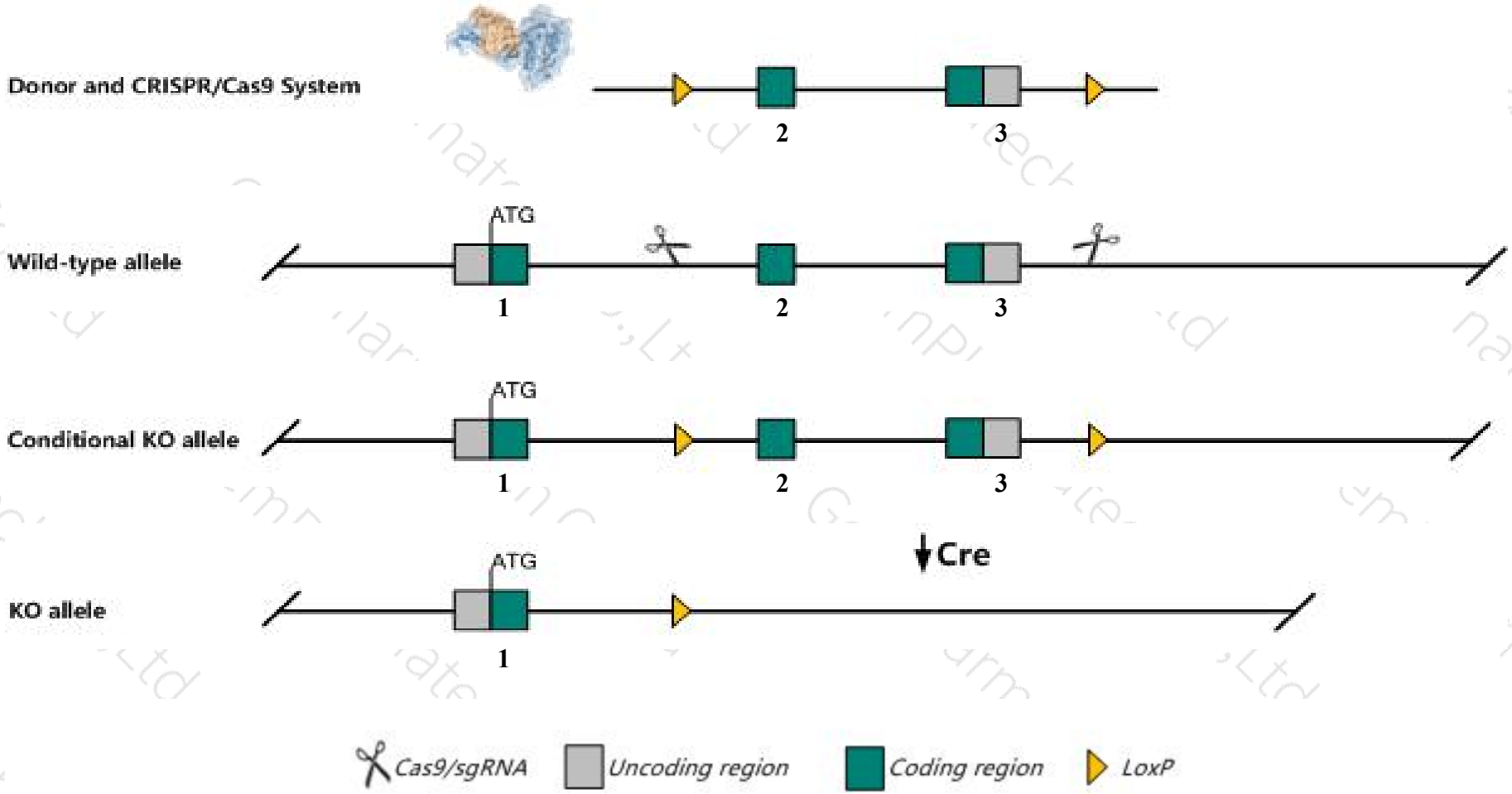
Project Overview

Project Name	***Htr2a***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Htr2a* gene. The schematic diagram is as follows:



Technical routes

- The *Htr2a* gene has 1 transcript. According to the structure of *Htr2a* gene, exon2-exon3 of *Htr2a-201* (ENSMUST00000036653.4) transcript is recommended as the knockout region.The region contains 1004bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Htr2a* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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,Mice homozygous for a knock-out allele show altered anxiety-related responses and increased vertical activity. Mice homozygous for a different knock-out allele exhibit abnormal enterocyte, Paneth cell and smooth muscle morphology.

- The *Htr2a* gene is located on the Chr14. 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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Htr2a 5-hydroxytryptamine (serotonin) receptor 2A [Mus musculus (house mouse)]

Gene ID: 15558, updated on 5-Mar-2019

Summary

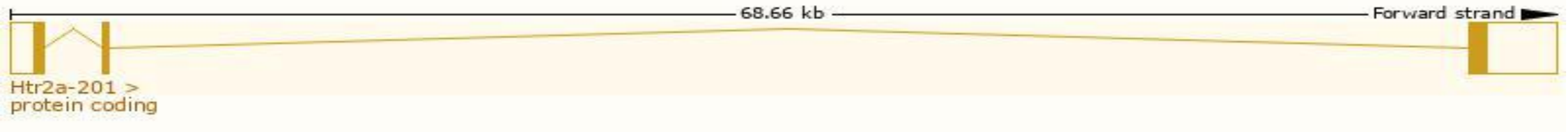
Official Symbol	Htr2a provided by MGI
Official Full Name	5-hydroxytryptamine (serotonin) receptor 2A provided by MGI
Primary source	MGI:MGI:109521
See related	Ensembl:ENSMUSG00000034997
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5-HT-2, 5-HT-2A, E030013E04, Htr-2, Htr2
Expression	Biased expression in frontal lobe adult (RPKM 3.5), cortex adult (RPKM 2.6) and 13 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 1 transcript,and the transcript is shown below:

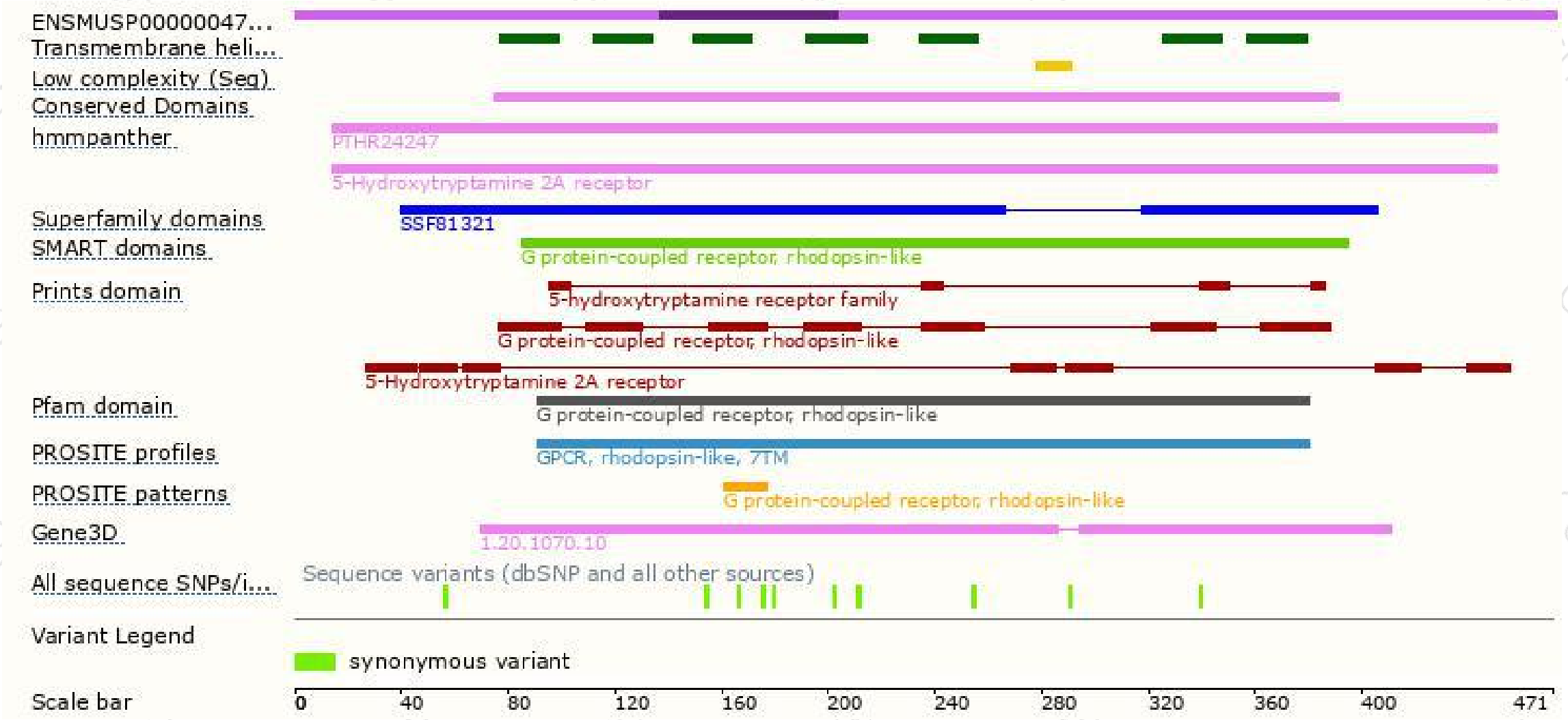
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Htr2a-201	ENSMUST00000036653.4	5606	471aa	Protein coding	CCDS27275	P35363 Q543D4	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of *Htr2a-201* transcript,The transcription is shown below

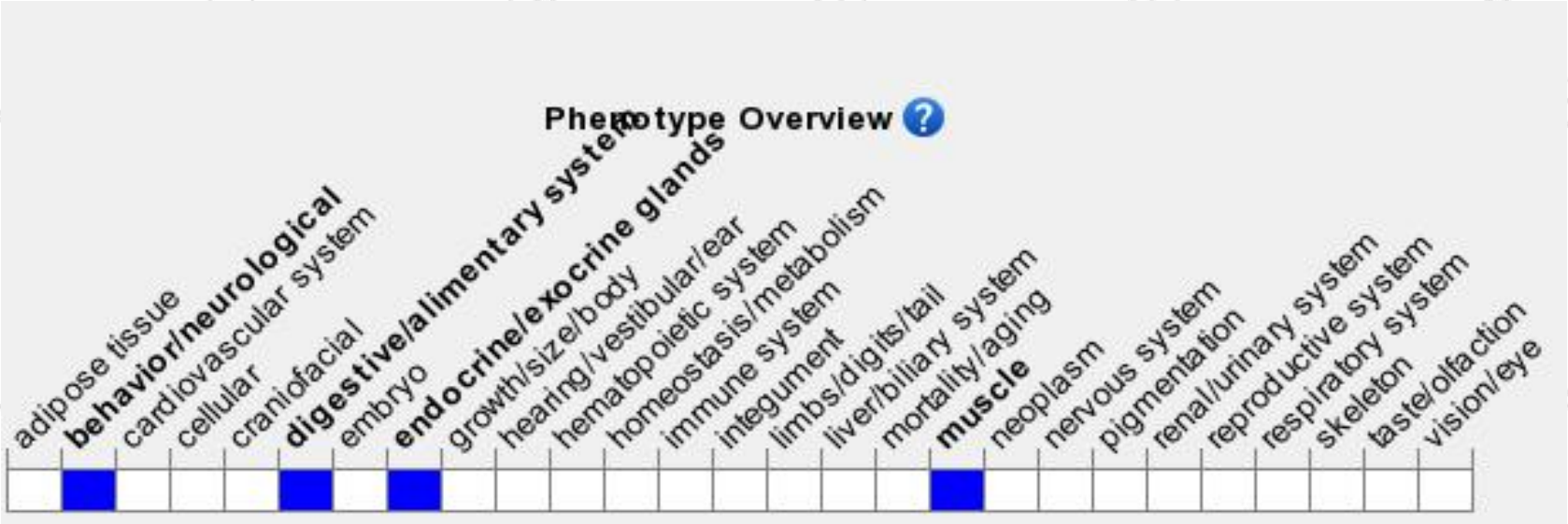


Genomic location distribution

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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