Trpc3 Cas9-CKO Strategy

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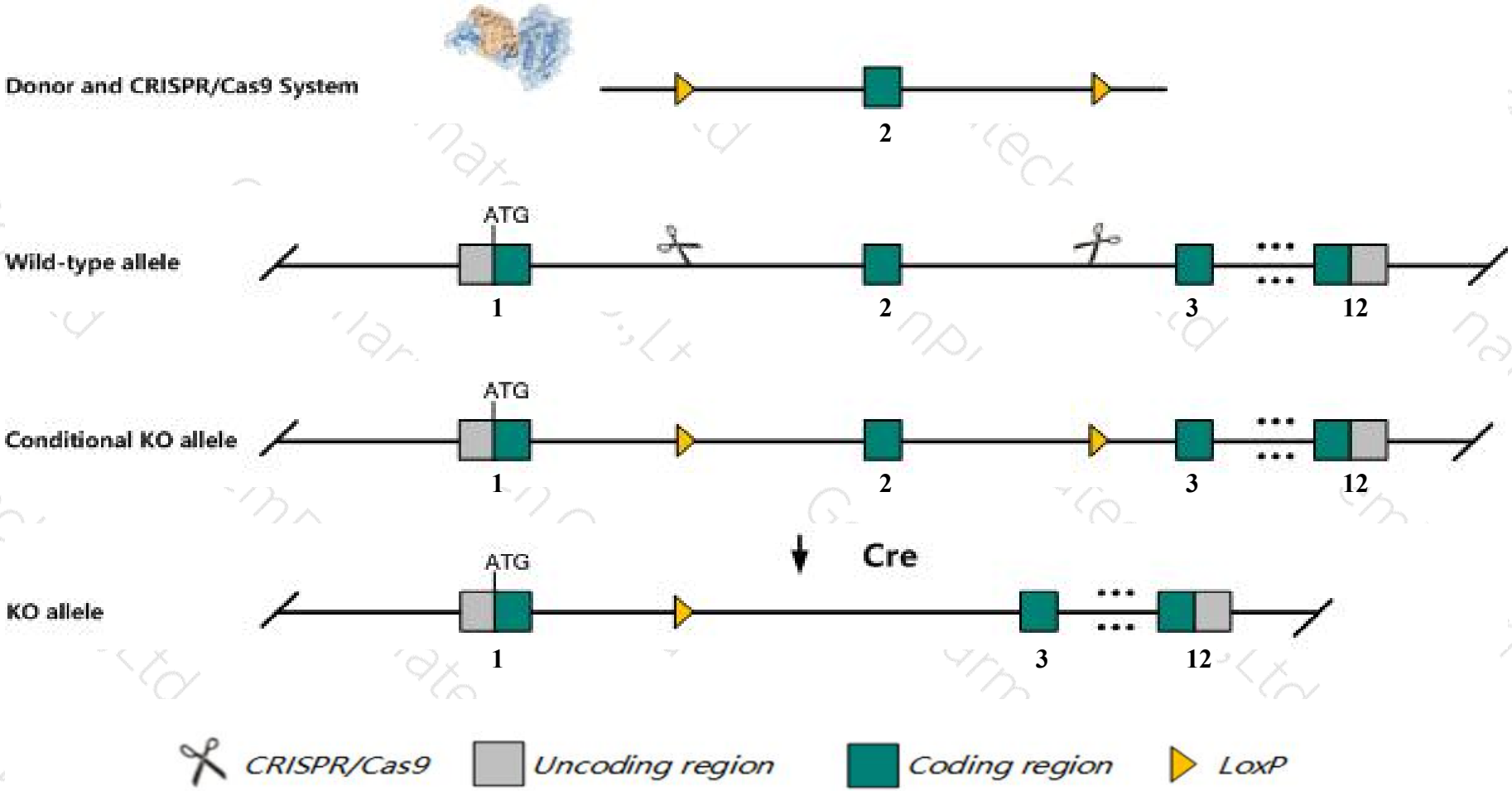
Project Overview

Project Name	***Trpc3***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Trpc3* gene. The schematic diagram is as follows:



Technical routes

- The *Trpc3* gene has 5 transcripts. According to the structure of *Trpc3* gene, exon2 of *Trpc3-201* (ENSMUST00000029271.4) transcript is recommended as the knockout region.The region contains 772bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Trpc3* 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,Homozygous knockout mice or mice heterozygoous for a point mutation in exon 7 display an abnormal gait. Abnormal nervous system electrophysiology is also described. An A1903G point mutation in exon 7 results in homozygous lethality.

➢ The *Trpc3* 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）

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Trpc3 transient receptor potential cation channel, subfamily C, member 3 [*Mus musculus* (house mouse)]

Gene ID: 22065, updated on 27-Aug-2019

Summary

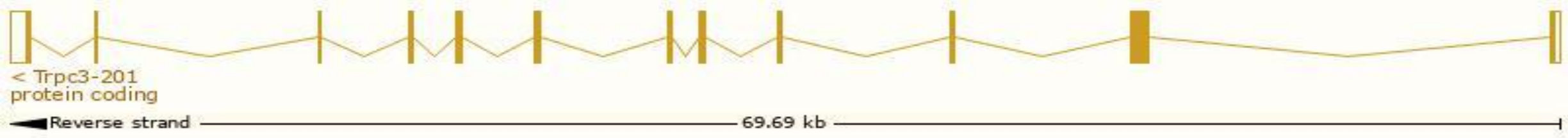
Official Symbol	Trpc3 provided by MGI
Official Full Name	transient receptor potential cation channel, subfamily C, member 3 provided by MGI
Primary source	MGI:MGI:109526
See related	Ensembl:ENSMUSG00000027716
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Mwk; Trp3; Trcp3; Trrp3
Expression	Biased expression in cerebellum adult (RPKM 16.9), CNS E18 (RPKM 4.1) and 11 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 5 transcripts,all transcripts are shown below:

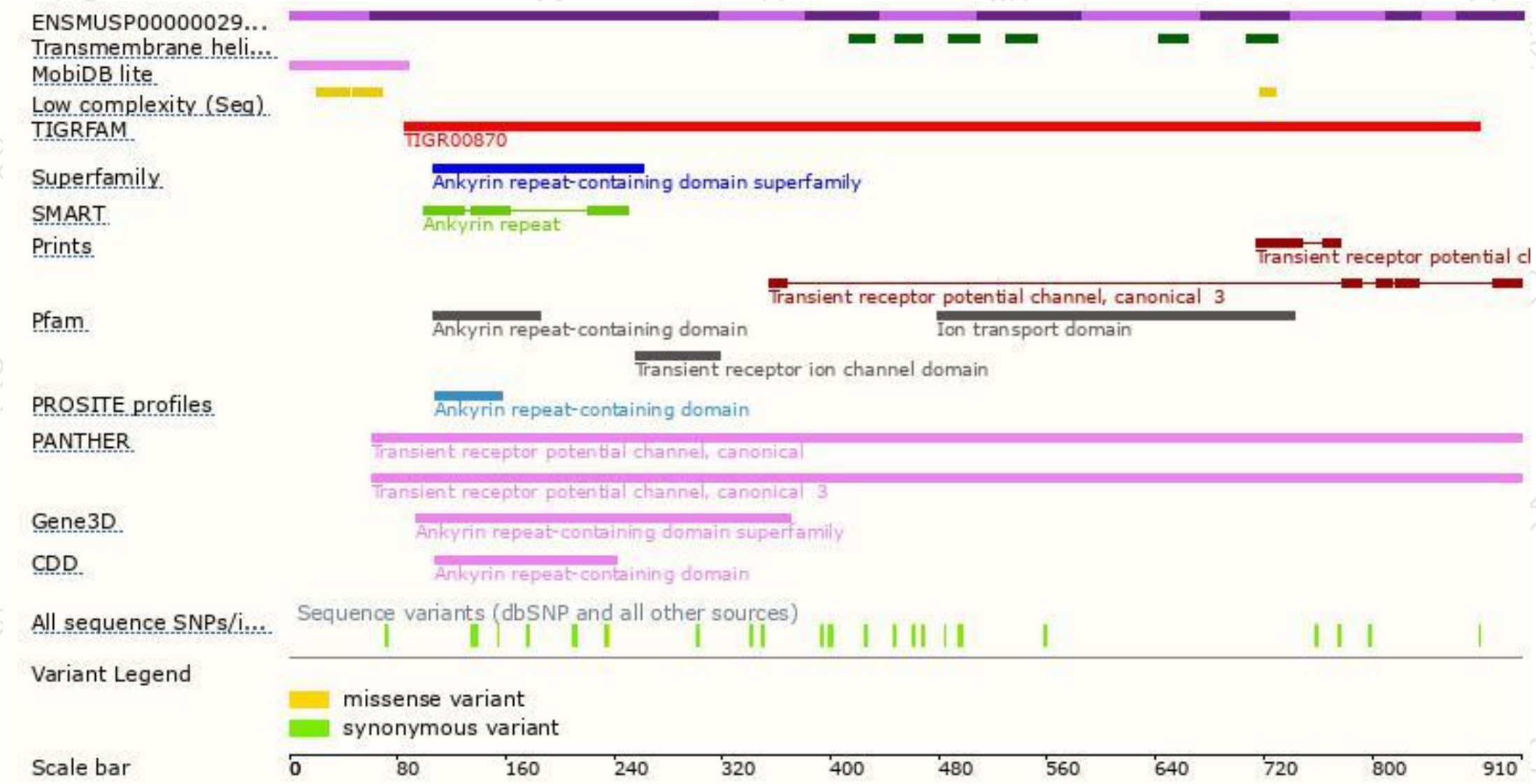
Name	Transcript ID	bp	Protein	Translation ID	Biotype	CCDS	UniProt	Flags
Trpc3-201	ENSMUST00000029271.4	3694	910aa	ENSMUSP00000029271.4	Protein coding	CCDS50893	B1ATV3	TSL:1 GENCODE basic APPRIS P1
Trpc3-205	ENSMUST00000146475.1	2811	No protein	-	Retained intron	-	-	TSL:1
Trpc3-204	ENSMUST00000133542.1	303	No protein	-	Retained intron	-	-	TSL:3
Trpc3-202	ENSMUST00000123220.1	361	No protein	-	lncRNA	-	-	TSL:3
Trpc3-203	ENSMUST00000129322.7	273	No protein	-	lncRNA	-	-	TSL:5

The strategy is based on the design of *Trpc3-201* transcript,The transcription is shown below

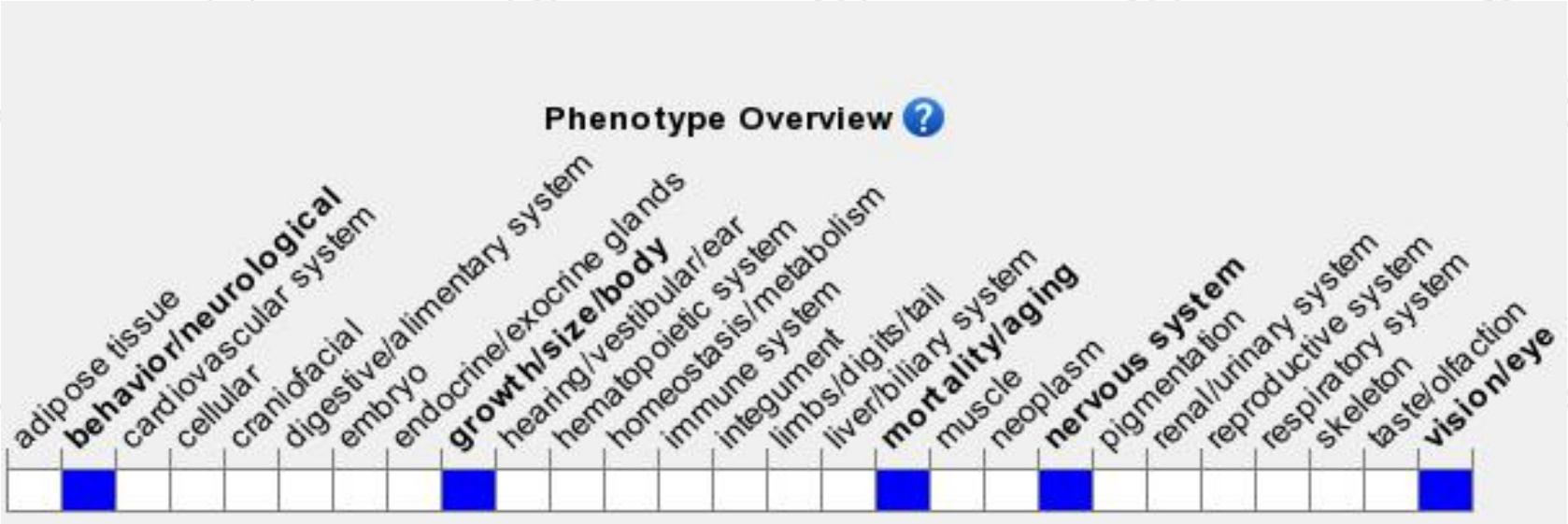


Genomic location distribution

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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