Cbfa2t2 Cas9-CKO Strategy

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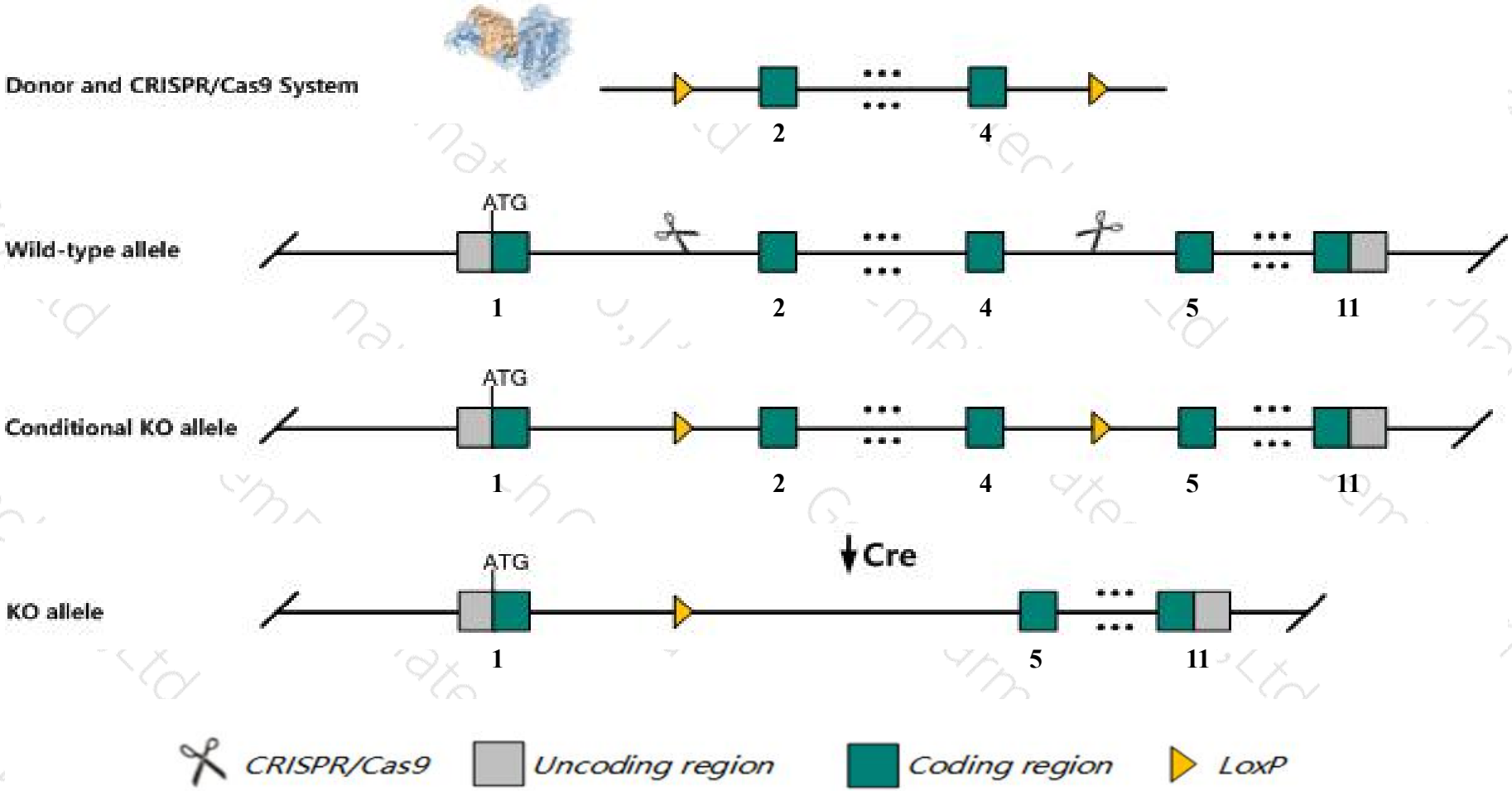
Design Date: 2020-5-28

Project Overview

Project Name	***Cbfa2t2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cbfa2t2* gene. The schematic diagram is as follows:



Technical routes

- The *Cbfa2t2* gene has 9 transcripts. According to the structure of *Cbfa2t2* gene, exon2-exon4 of *Cbfa2t2-201* (ENSMUST00000045270.14) transcript is recommended as the knockout region.The region contains 476bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cbfa2t2* 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 a null allele are smaller and show reduced numbers of intestinal goblet, paneth and enteroendocrine cells, small intestine inflammation, and strain dependent postnatal lethality. homozygotes for a different null allele are infertile due to defects in primordial germ cell maturation.
- The *Cbfa2t2* gene is located on the Chr2. 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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Cbfa2t2 CBFA2/RUNX1 translocation partner 2 [Mus musculus (house mouse)]

Gene ID: 12396, updated on 13-Mar-2020

Summary

Official Symbol	Cbfa2t2 provided by MGI
Official Full Name	CBFA2/RUNX1 translocation partner 2 provided byMGI
Primary source	MGI:MGI:1333833
See related	Ensembl:ENSMUSG00000038533
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	A430091M07, C330013D05Rik, Cbfa2t2h, MTGR1
Expression	Ubiquitous expression in whole brain E14.5 (RPKM 10.6), CNS E14 (RPKM 10.3) and 28 other tissuesSee more
Orthologs	human all

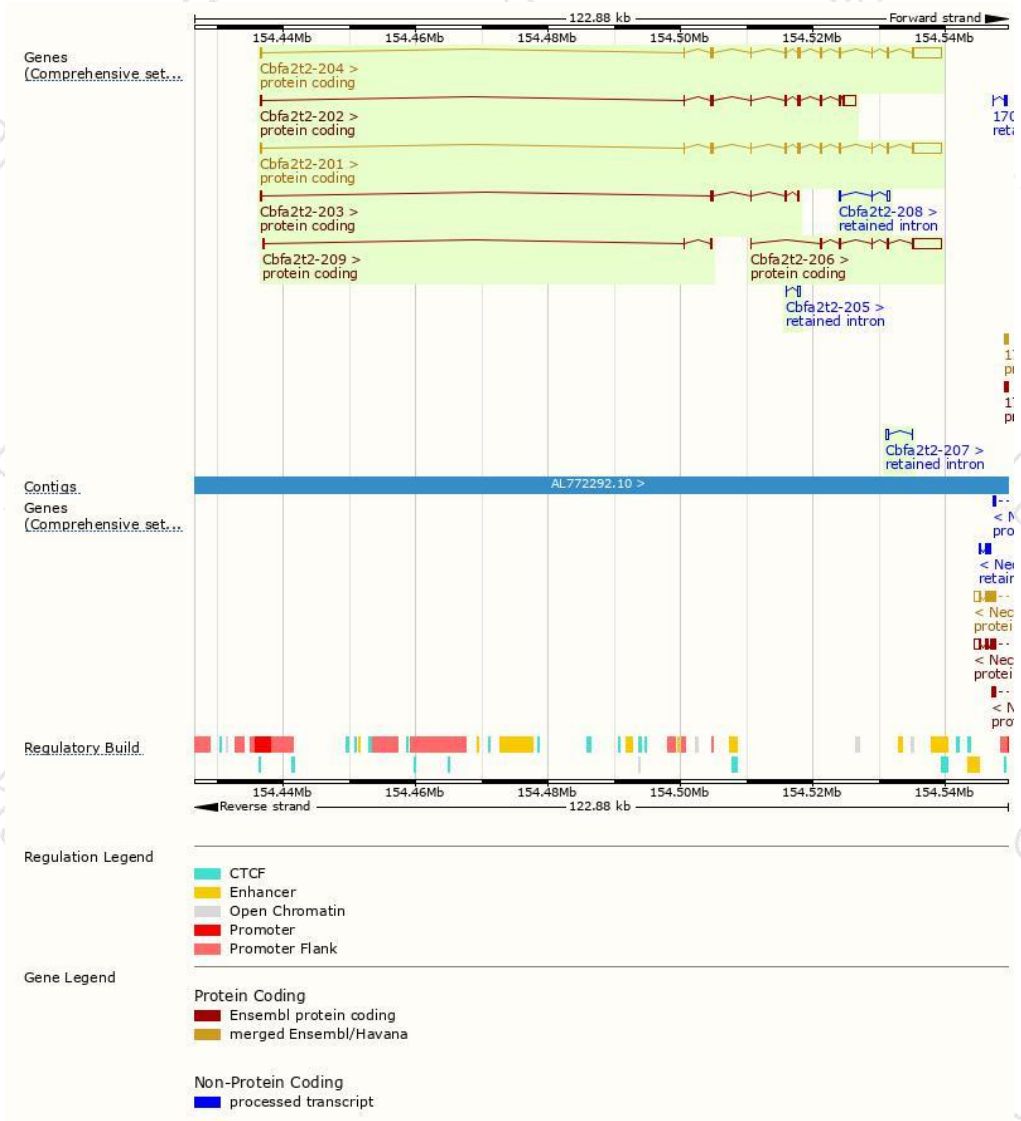
Transcript information（Ensembl）

The gene has 9 transcripts,all transcripts are shown below:

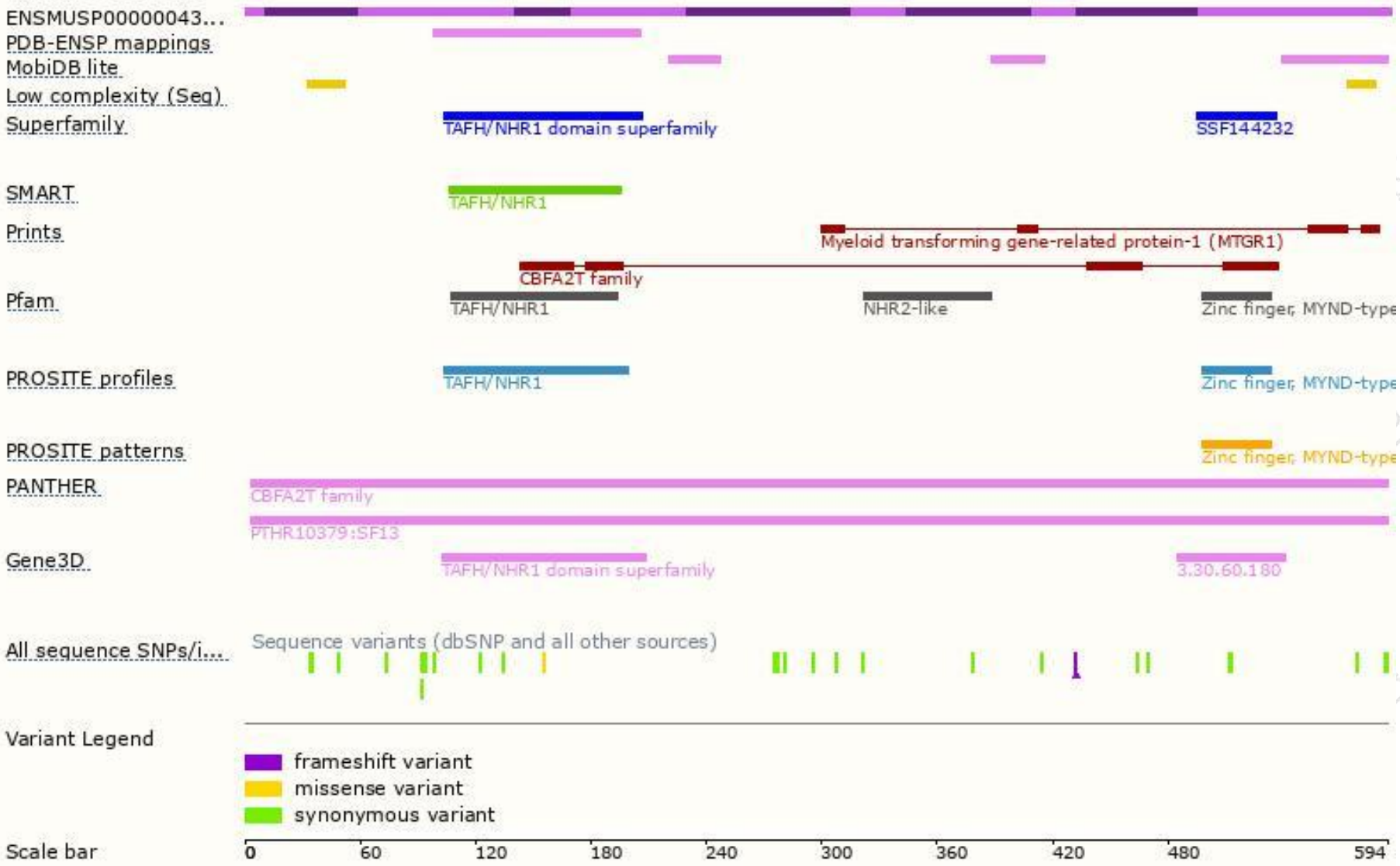
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cbfa2t2-204	ENSMUST00000109725.7	6080	593aa	Protein coding	CCDS50764	A0A0R4J1D5	TSL:1 GENCODE basic APPRIS ALT1
Cbfa2t2-201	ENSMUST00000045270.14	6069	594aa	Protein coding	CCDS16932	O70374	TSL:1 GENCODE basic APPRIS P3
Cbfa2t2-206	ENSMUST00000137526.1	5074	299aa	Protein coding	-	F6WYU8	CDS 5' incomplete TSL:3
Cbfa2t2-202	ENSMUST00000099178.9	3247	473aa	Protein coding	-	Q3UGB2	TSL:1 GENCODE basic
Cbfa2t2-203	ENSMUST00000109724.1	732	221aa	Protein coding	-	A2AKD9	CDS 3' incomplete TSL:3
Cbfa2t2-209	ENSMUST00000155202.1	576	192aa	Protein coding	-	F6QMQ3	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Cbfa2t2-208	ENSMUST00000154487.1	560	No protein	Retained intron	-	-	TSL:3
Cbfa2t2-207	ENSMUST00000139506.1	455	No protein	Retained intron	-	-	TSL:2
Cbfa2t2-205	ENSMUST00000135647.1	381	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Cbfa2t2-201* transcript,the transcription is shown below:

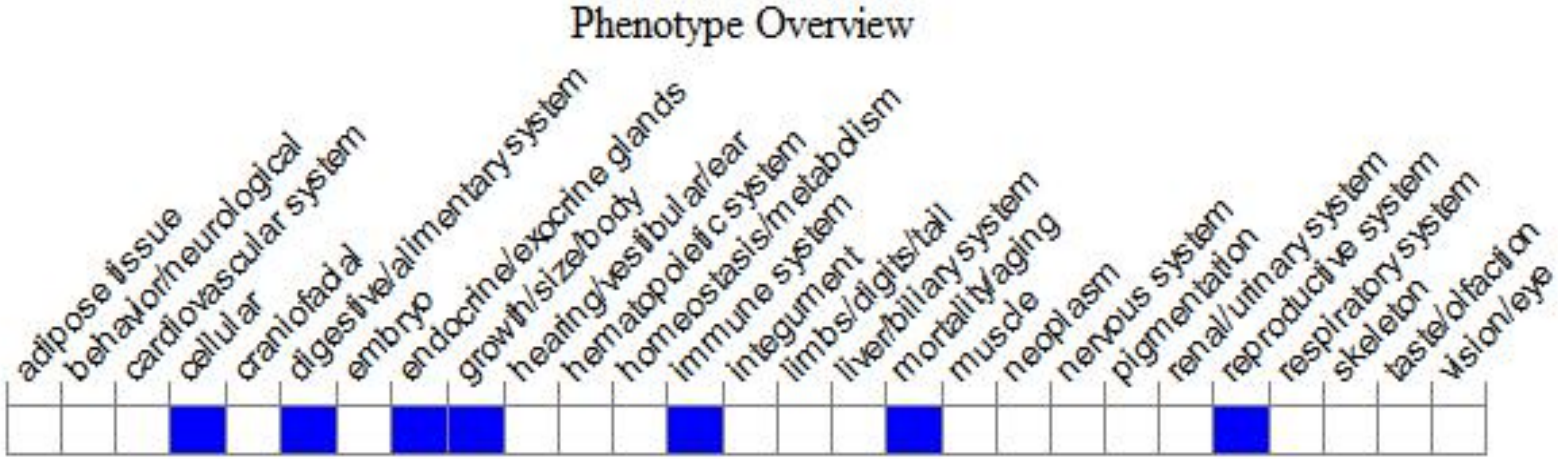
Genomic location distribution



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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