Dnajb4 Cas9-CKO Strategy

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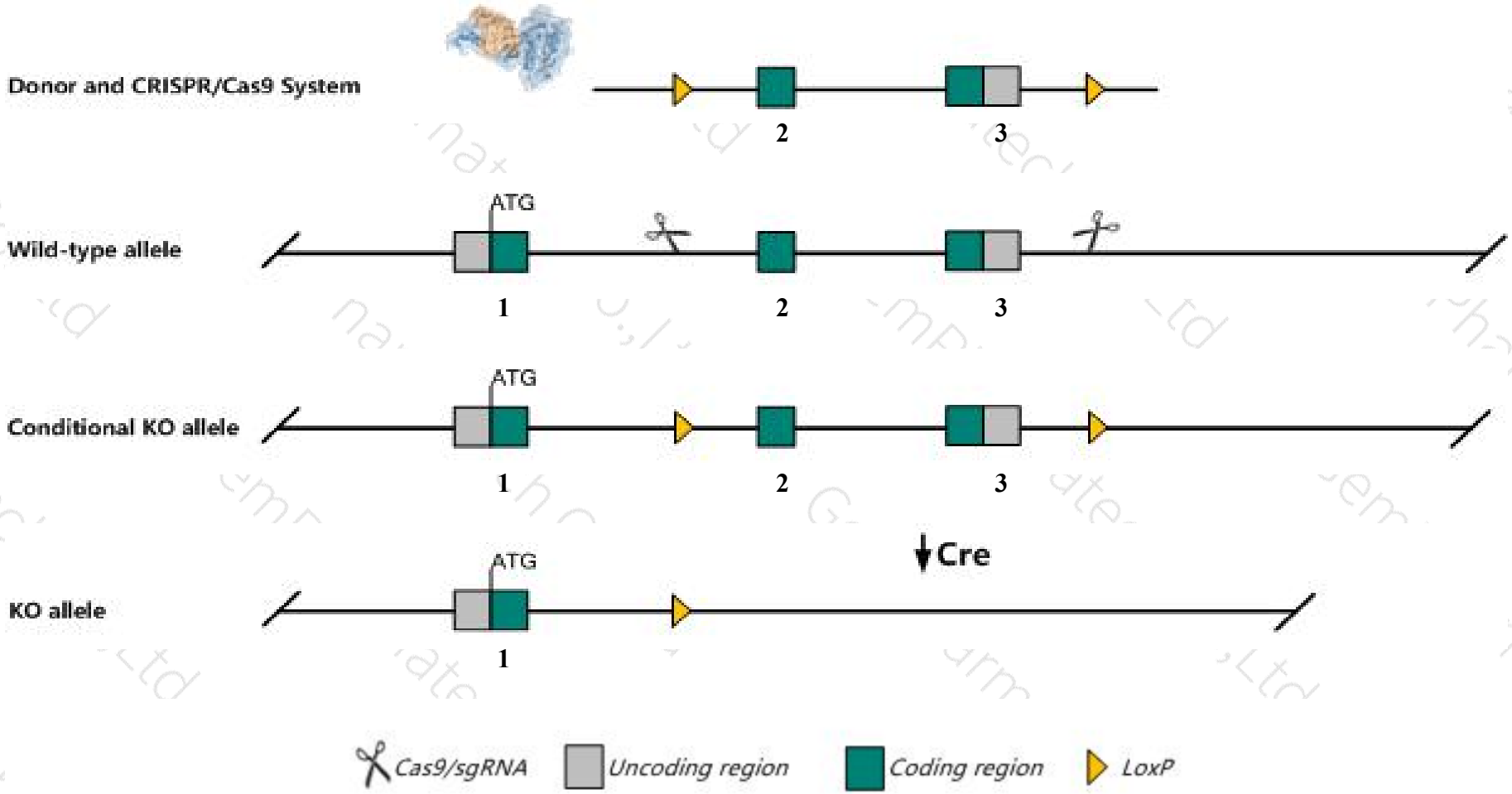
Project Overview

Project Name	***Dnajb4***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Dnajb4* gene. The schematic diagram is as follows:



Technical routes

➢ The *Dnajb4* gene has 7 transcripts. According to the structure of *Dnajb4* gene, exon2-exon3 of *Dnajb4-202*(ENSMUST00000050073.12) transcript is recommended as the knockout region.The region contains 803bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Dnajb4* 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

➢ The *Dnajb4* 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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Dnajb4 DnaJ heat shock protein family (Hsp40) member B4 [Mus musculus (house mouse)]

Gene ID: 67035, updated on 13-Mar-2020

Summary

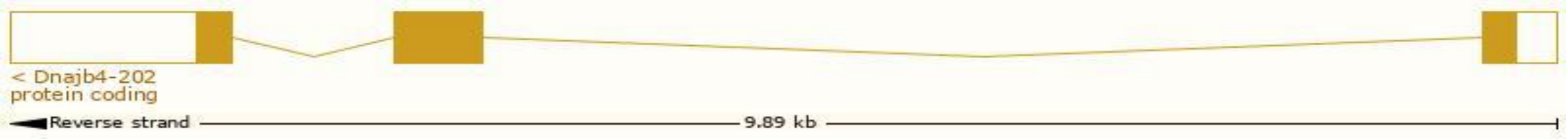
Official Symbol	Dnajb4 provided by MGI
Official Full Name	DnaJ heat shock protein family (Hsp40) member B4 provided byMGI
Primary source	MGI:MGI:1914285
See related	Ensembl:ENSMUSG00000028035
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1700029A20Rik, 2010306G19Rik, 5730460G06Rik
Expression	Broad expression in testis adult (RPKM 16.1), bladder adult (RPKM 10.9) and 24 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

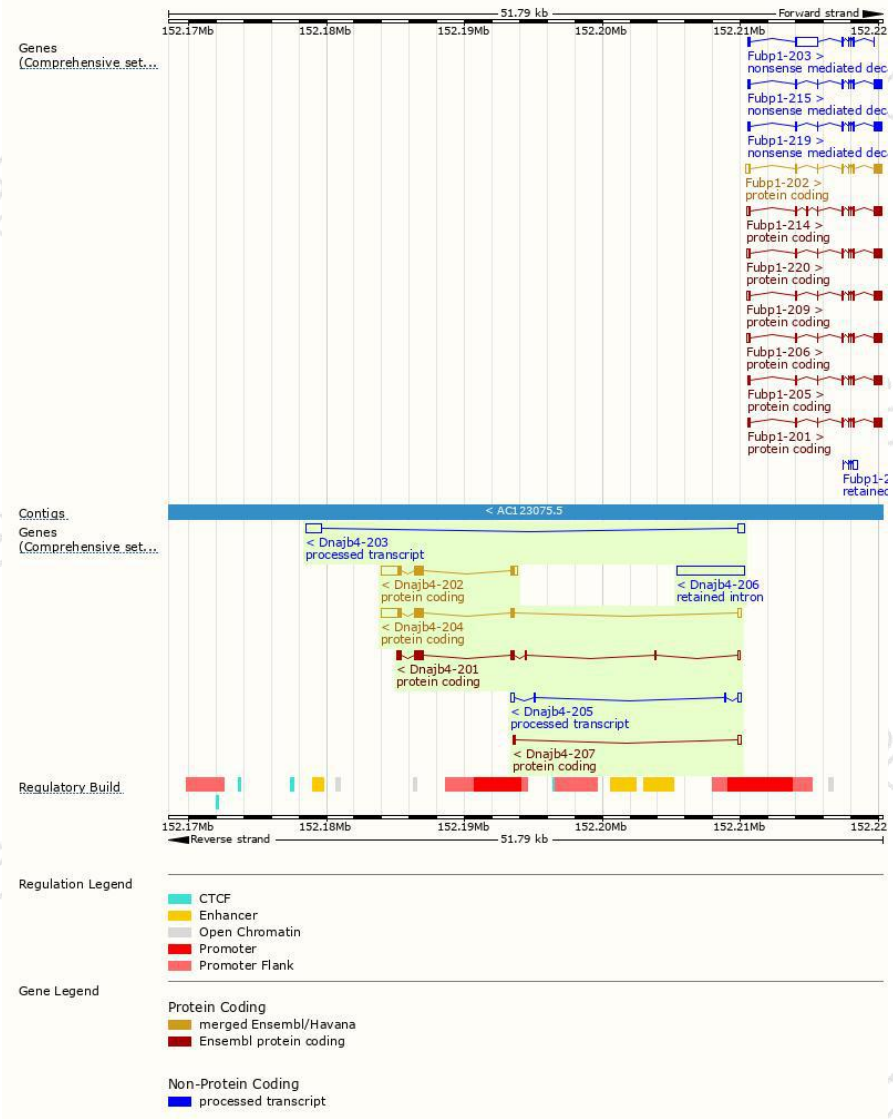
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dnajb4-202	ENSMUST00000050073.12	2464	337aa	Protein coding	CCDS17915	Q9D832	TSL:1 GENCODE basic APPRIS P1
Dnajb4-204	ENSMUST00000144950.7	2459	337aa	Protein coding	CCDS17915	Q9D832	TSL:1 GENCODE basic APPRIS P1
Dnajb4-201	ENSMUST00000029669.3	1497	337aa	Protein coding	CCDS17915	Q9D832	TSL:5 GENCODE basic APPRIS P1
Dnajb4-207	ENSMUST00000197941.1	325	8aa	Protein coding	-	A0A0G2JEM9	CDS 3' incomplete TSL:3
Dnajb4-203	ENSMUST00000134701.1	1581	No protein	Processed transcript	-	-	TSL:1
Dnajb4-205	ENSMUST00000153355.5	579	No protein	Processed transcript	-	-	TSL:3
Dnajb4-206	ENSMUST00000196291.1	4913	No protein	Retained intron	-	-	TSL:NA

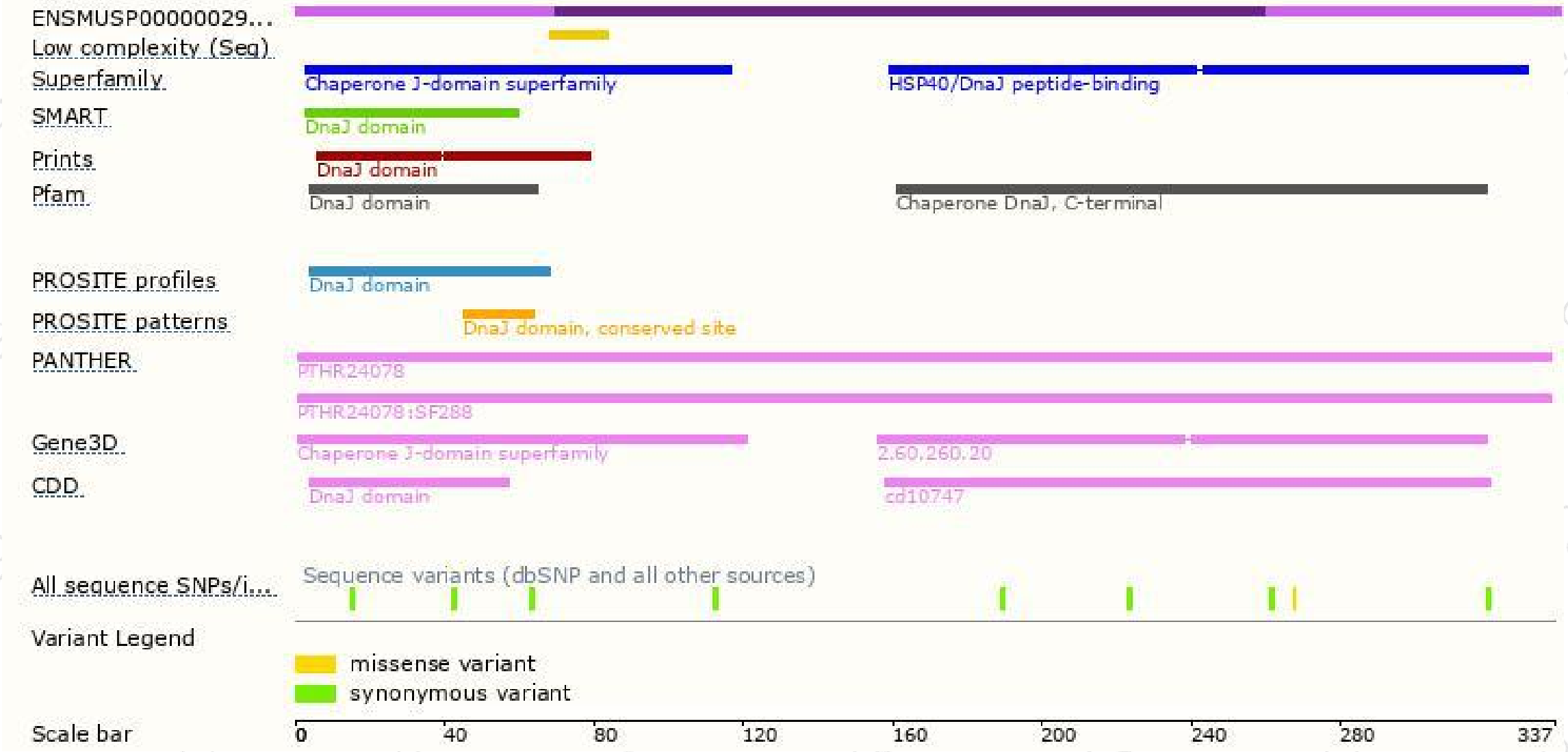
The strategy is based on the design of *Dnajb4-202* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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