

***Dtnb* Cas9-CKO Strategy**

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Project Overview

Project Name

Dtnb

Project type

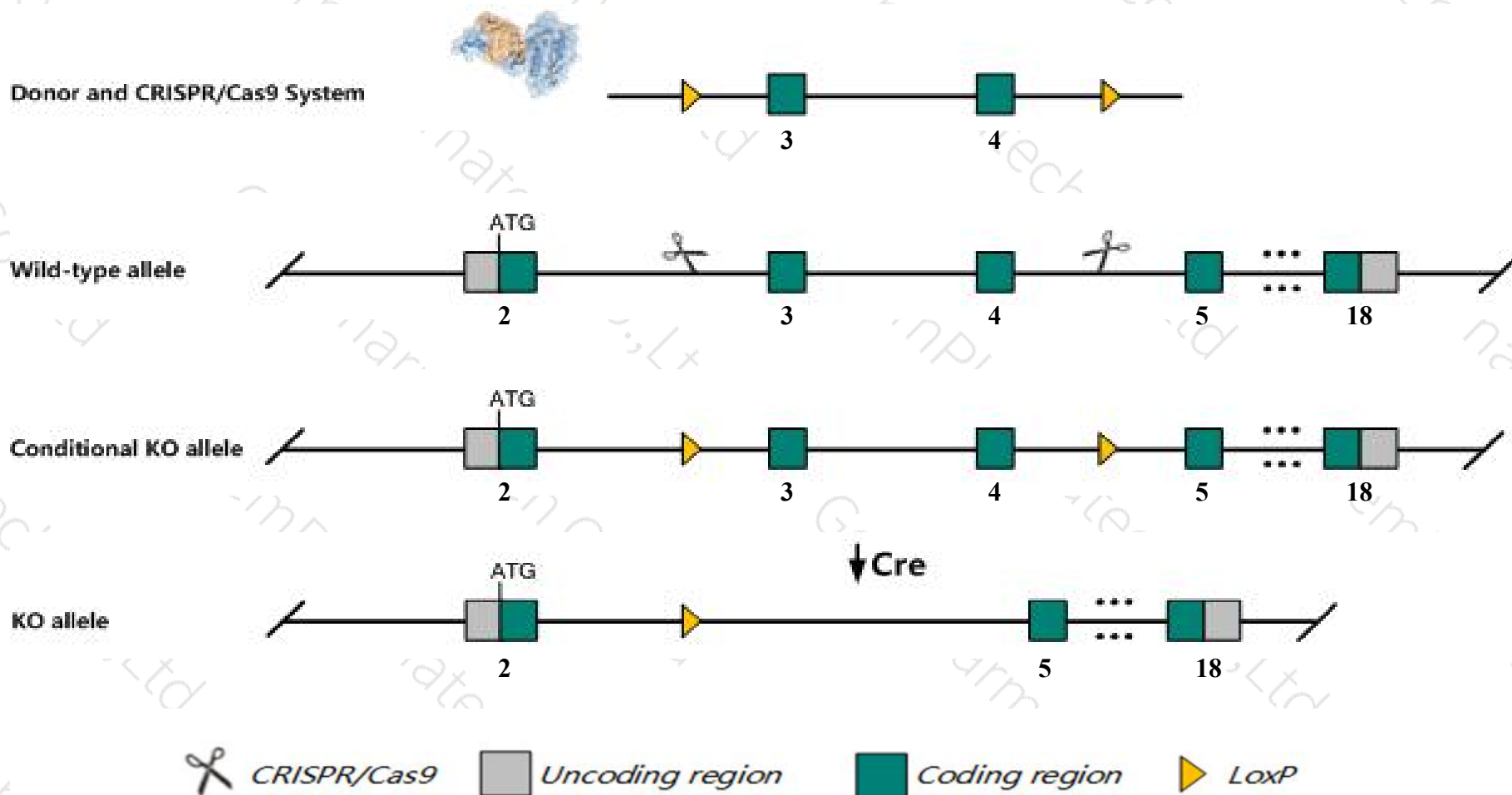
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Dtnb* gene has 25 transcripts. According to the structure of *Dtnb* gene, exon3-exon4 of *Dtnb*-203 (ENSMUST00000164578.8) transcript is recommended as the knockout region. The region contains 295bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Dtnb* 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.

- According to the existing MGI data, Mice homozygous for a knock-out allele are viable and fertile and show no obvious histological or functional abnormalities in liver and kidney. Mice homozygous for a gene trapped allele are viable, fertile and overtly normal with no significant synaptic or behavioral defects.
- The *Dtnb* gene is located on the Chr12. 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)

Dtnb dystrobrevin, beta [*Mus musculus* (house mouse)]

Gene ID: 13528, updated on 24-Oct-2019

Summary

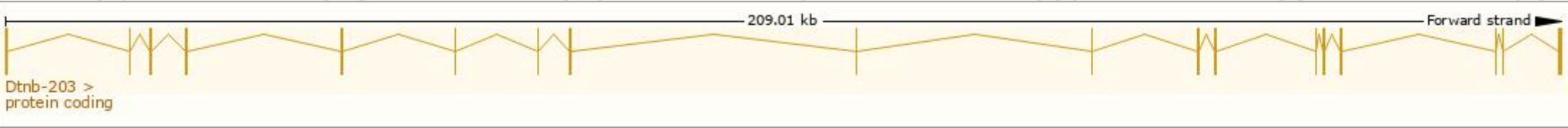
Official Symbol	Dtnb provided by MGI
Official Full Name	dystrobrevin, beta provided by MGI
Primary source	MGI:MGI:1203728
See related	Ensembl:ENSMUSG000000071454
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Ubiquitous expression in testis adult (RPKM 19.5), frontal lobe adult (RPKM 9.3) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

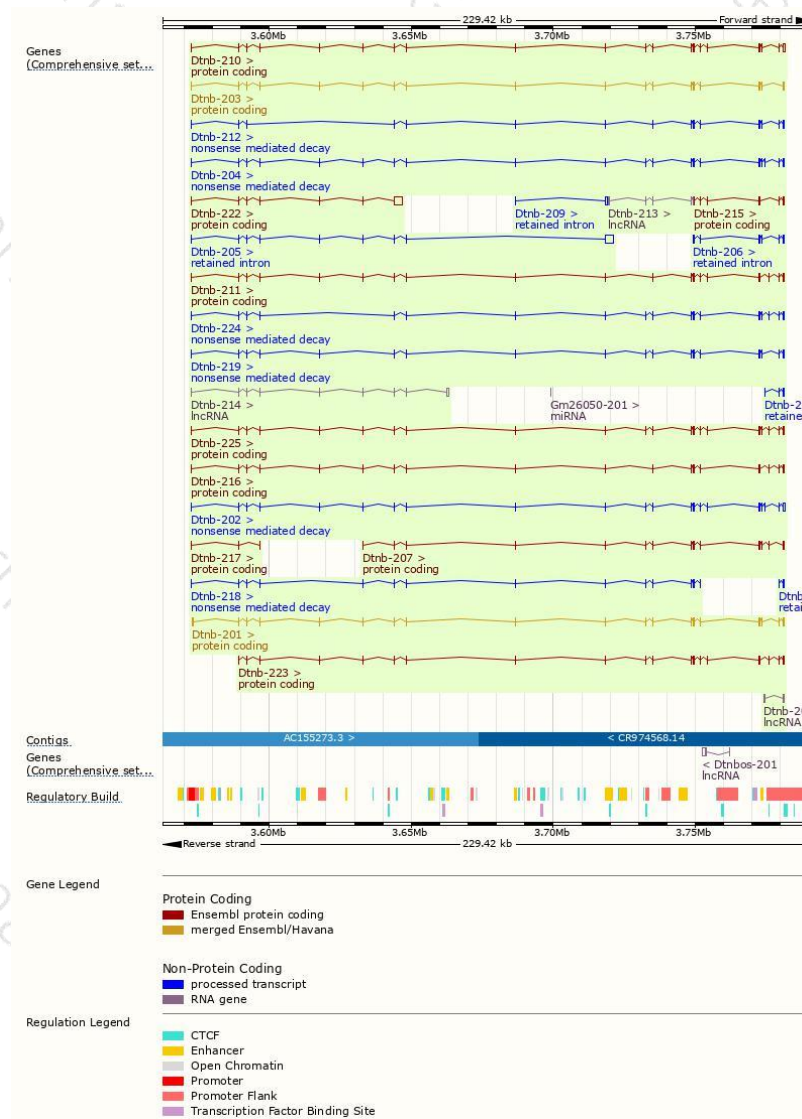
The gene has 25 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dtnb-201	ENSMUST00000077930.12	2376	602aa	Protein coding	CCDS36396	Q8K0N0	TSL1 GENCODE basic APPRIS P2
Dtnb-203	ENSMUST00000164578.8	2355	659aa	Protein coding	CCDS49015	Q70585	TSL1 GENCODE basic
Dtnb-222	ENSMUST00000174479.7	3689	265aa	Protein coding	-	G3UXI3	TSL1 GENCODE basic
Dtnb-210	ENSMUST00000173199.7	2820	609aa	Protein coding	-	G3UZ87	TSL5 GENCODE basic APPRIS ALT1
Dtnb-225	ENSMUST00000174663.7	2381	626aa	Protein coding	-	G3UYM7	TSL5 GENCODE basic APPRIS ALT1
Dtnb-211	ENSMUST00000173240.7	2218	566aa	Protein coding	-	G3UXK0	TSL5 GENCODE basic
Dtnb-216	ENSMUST00000173736.7	2175	559aa	Protein coding	-	G3UZJ3	TSL5 GENCODE basic
Dtnb-223	ENSMUST00000174547.7	2112	596aa	Protein coding	-	G3UY34	TSL5 GENCODE basic
Dtnb-207	ENSMUST00000172598.7	1585	490aa	Protein coding	-	G3UYK9	CDS 5' incomplete TSL5
Dtnb-215	ENSMUST00000173542.7	648	107aa	Protein coding	-	G3UYD1	CDS 5' incomplete TSL3
Dtnb-217	ENSMUST00000173998.7	384	90aa	Protein coding	-	G3UXP7	CDS 3' incomplete TSL3
Dtnb-202	ENSMUST00000101637.10	2428	615aa	Nonsense mediated decay	-	Q70585	TSL1
Dtnb-204	ENSMUST00000164607.8	2390	608aa	Nonsense mediated decay	-	Q8C8Z9	TSL1
Dtnb-219	ENSMUST00000174290.7	2157	149aa	Nonsense mediated decay	-	G3UXH9	TSL5
Dtnb-224	ENSMUST00000174639.7	1873	123aa	Nonsense mediated decay	-	G3UX96	TSL5
Dtnb-212	ENSMUST00000173483.7	1781	63aa	Nonsense mediated decay	-	G3UZ85	TSL5
Dtnb-218	ENSMUST00000174109.7	1493	138aa	Nonsense mediated decay	-	G3UYN2	TSL5
Dtnb-205	ENSMUST00000168806.7	3768	No protein	Retained intron	-	-	TSL1
Dtnb-206	ENSMUST00000172504.7	715	No protein	Retained intron	-	-	TSL2
Dtnb-209	ENSMUST00000173074.1	664	No protein	Retained intron	-	-	TSL2
Dtnb-221	ENSMUST00000174414.1	414	No protein	Retained intron	-	-	TSL2
Dtnb-220	ENSMUST00000174293.1	400	No protein	Retained intron	-	-	TSL2
Dtnb-214	ENSMUST00000173524.7	1629	No protein	lncRNA	-	-	TSL1
Dtnb-208	ENSMUST00000172735.1	545	No protein	lncRNA	-	-	TSL3
Dtnb-213	ENSMUST00000173498.1	368	No protein	lncRNA	-	-	TSL5

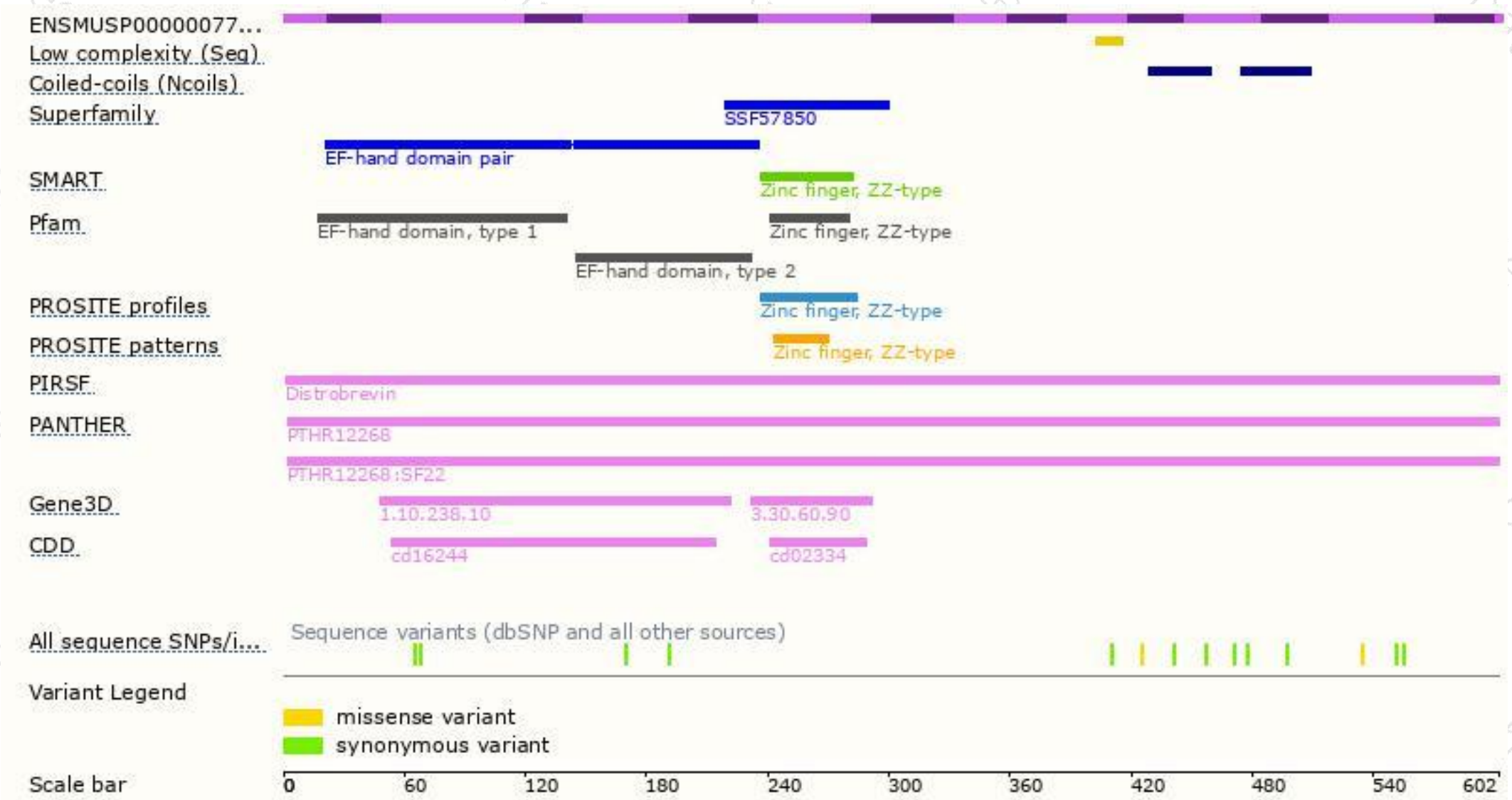
The strategy is based on the design of *Dtnb-203* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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