

Btbd2 Cas9-CKO Strategy

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Project Overview



Project Name

Project type Cas9-CKO

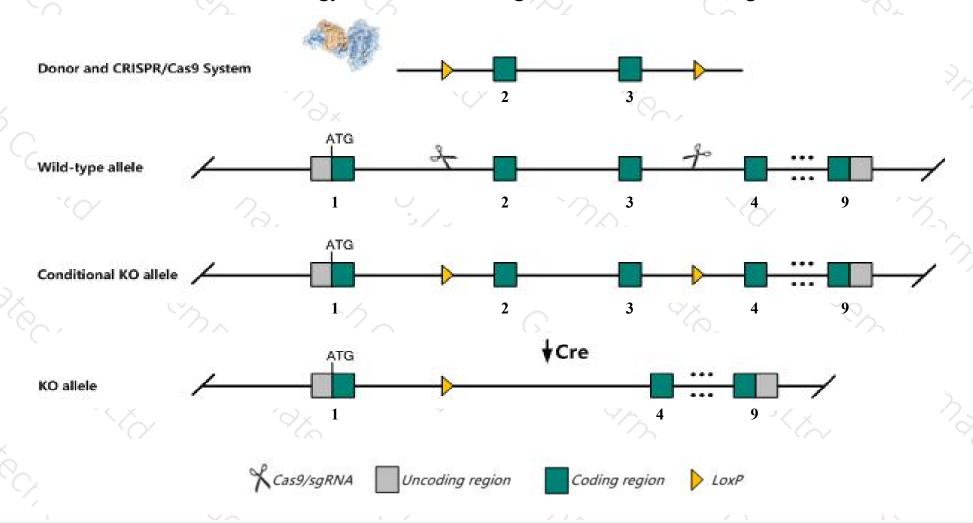
Strain background C57BL/6JGpt

Btbd2

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Btbd2* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Btbd2* gene has 5 transcripts. According to the structure of *Btbd2* gene, exon2-exon3 of *Btbd2*201(ENSMUST00000003434.13) transcript is recommended as the knockout region. The region contains 277bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Btbd2* 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 *Btbd2* gene is located on the Chr10. 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.
- ➤ Transcript *Btbd2*-204 may not be affected.
- 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)



Btbd2 BTB (POZ) domain containing 2 [Mus musculus (house mouse)]

Gene ID: 208198, updated on 23-Mar-2020

Summary

☆ ?

Official Symbol Btbd2 provided by MGI

Official Full Name BTB (POZ) domain containing 2 provided by MGI

Primary source MGI:MGI:1933831

See related Ensembl: ENSMUSG00000003344

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 2610037C03Rik, 4930512K17Rik

Expression Ubiquitous expression in adrenal adult (RPKM 45.6), ovary adult (RPKM 43.7) and 28 other tissuesSee more

Orthologs <u>human all</u>

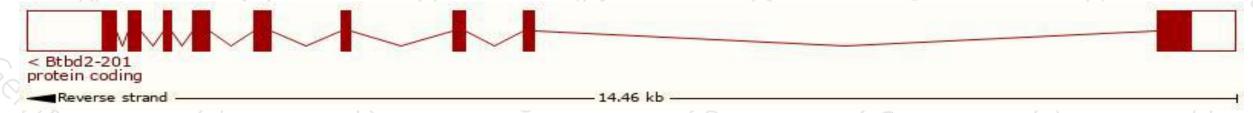
Transcript information (Ensembl)



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

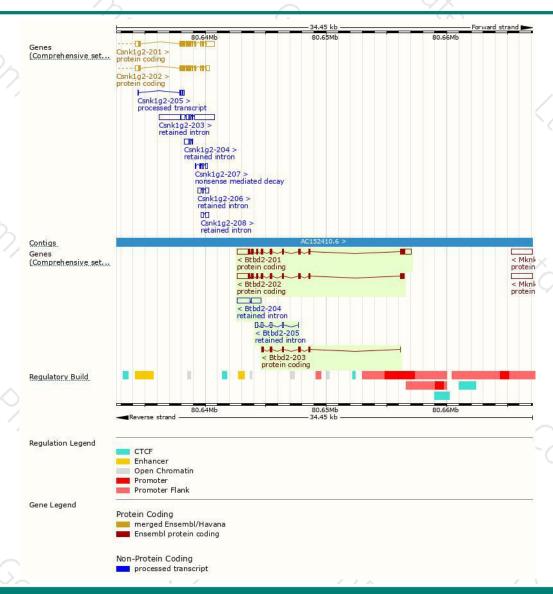
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Btbd2-201	ENSMUST00000003434.13	3026	523aa	Protein coding	CCDS48639	E9PUS2	TSL:5 GENCODE basic APPRIS P2
Btbd2-202	ENSMUST00000126980.7	2449	508aa	Protein coding		E9PY28	TSL:5 GENCODE basic APPRIS ALT2
Btbd2-203	ENSMUST00000131876.1	766	<u>256aa</u>	Protein coding	858	F6WBW4	CDS 5' and 3' incomplete TSL:5
Btbd2-204	ENSMUST00000147162.1	1876	No protein	Retained intron			TSL:1
Btbd2-205	ENSMUST00000151794.1	722	No protein	Retained intron	125	2	TSL:3

The strategy is based on the design of *Btbd2-201* transcript, the transcription is shown below:



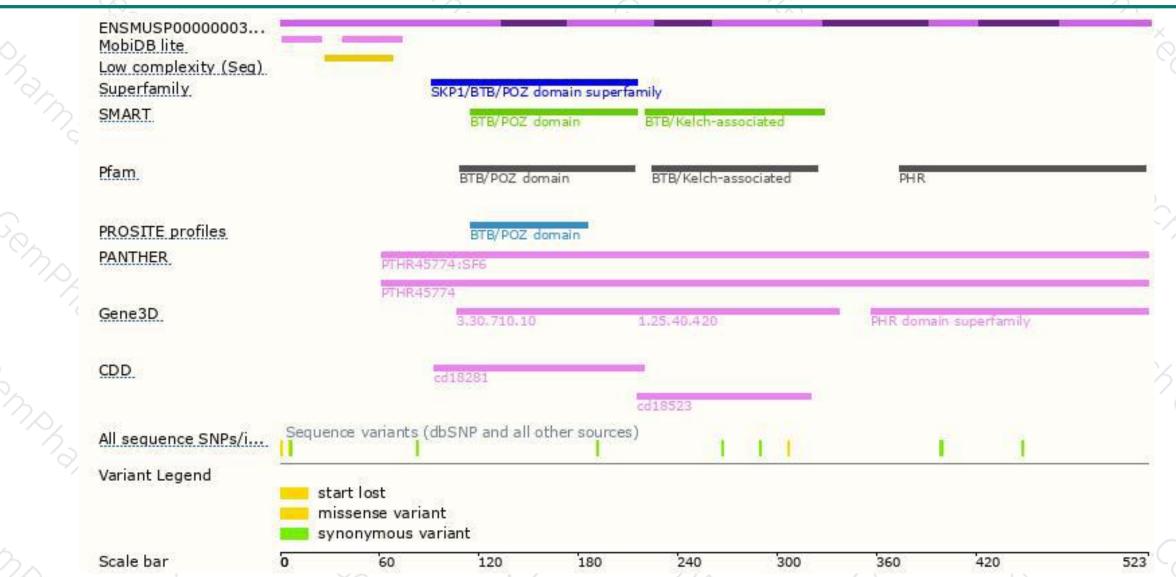
Genomic location distribution





Protein domain







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

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