

Fbxw2 Cas9-CKO Strategy

Designer: Shuang Zhang

Reviewer: Yun Li

Design Date: 2021-5-24

Project Overview

Project Name

Fbxw2

Project type

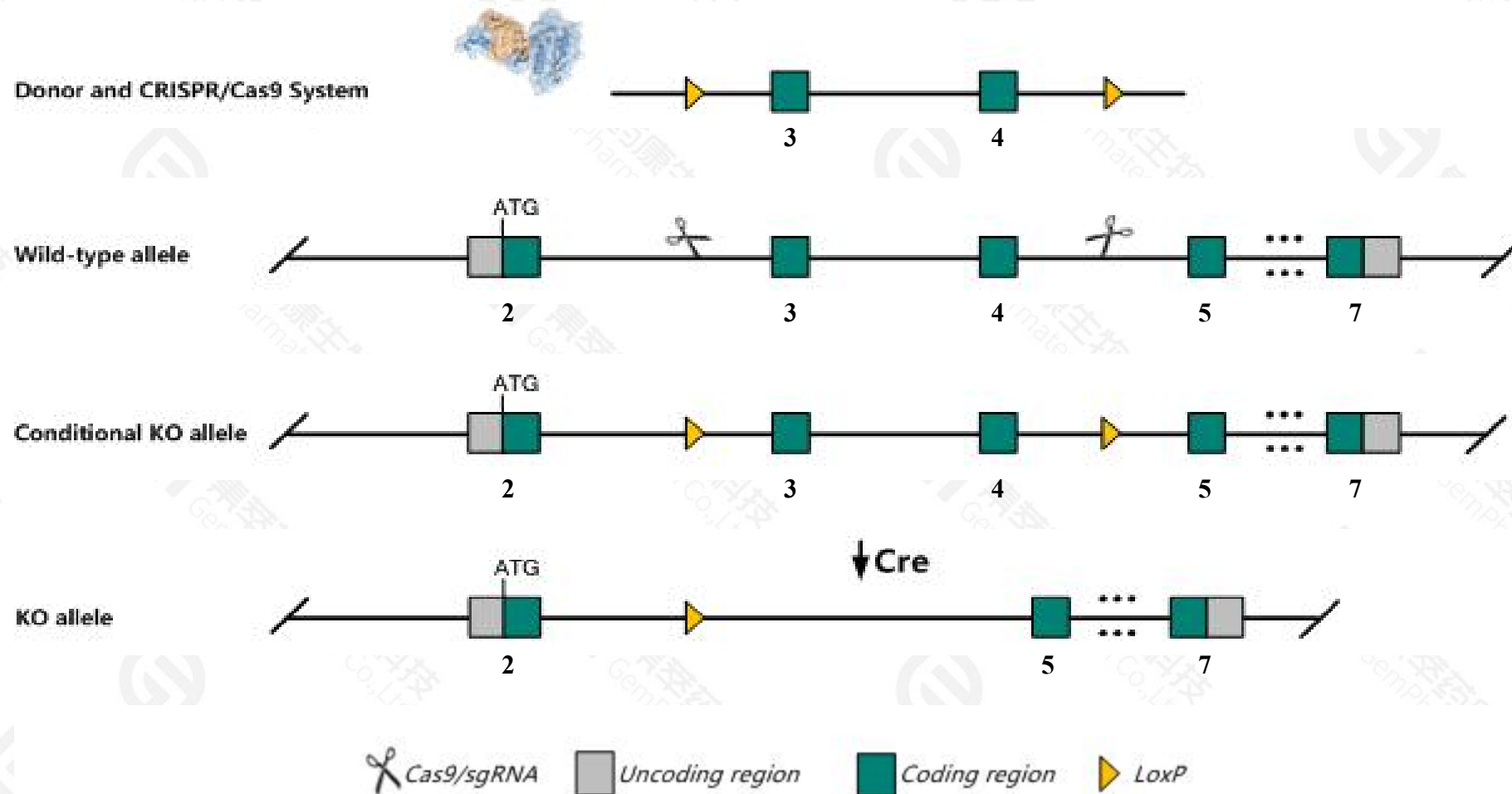
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Fbxw2* gene. The schematic diagram is as follows:



- The *Fbxw2* gene has 14 transcripts. According to the structure of *Fbxw2* gene, exon3-exon4 of *Fbxw2*-201(ENSMUST00000028220.10) transcript is recommended as the knockout region. The region contains 329bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Fbxw2* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor 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.

- The *Fbxw2* 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)

Fbxw2 F-box and WD-40 domain protein 2 [Mus musculus (house mouse)]

Gene ID: 30050, updated on 12-Feb-2021

Summary



Official Symbol	Fbxw2 provided by MGI
Official Full Name	F-box and WD-40 domain protein 2 provided by MGI
Primary source	MGI:MGI:1353435
See related	Ensembl:ENSMUSG00000035949
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	2700071L08Rik, FBW2, Fwd2, MD6
Expression	Ubiquitous expression in CNS E18 (RPKM 8.5), bladder adult (RPKM 8.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

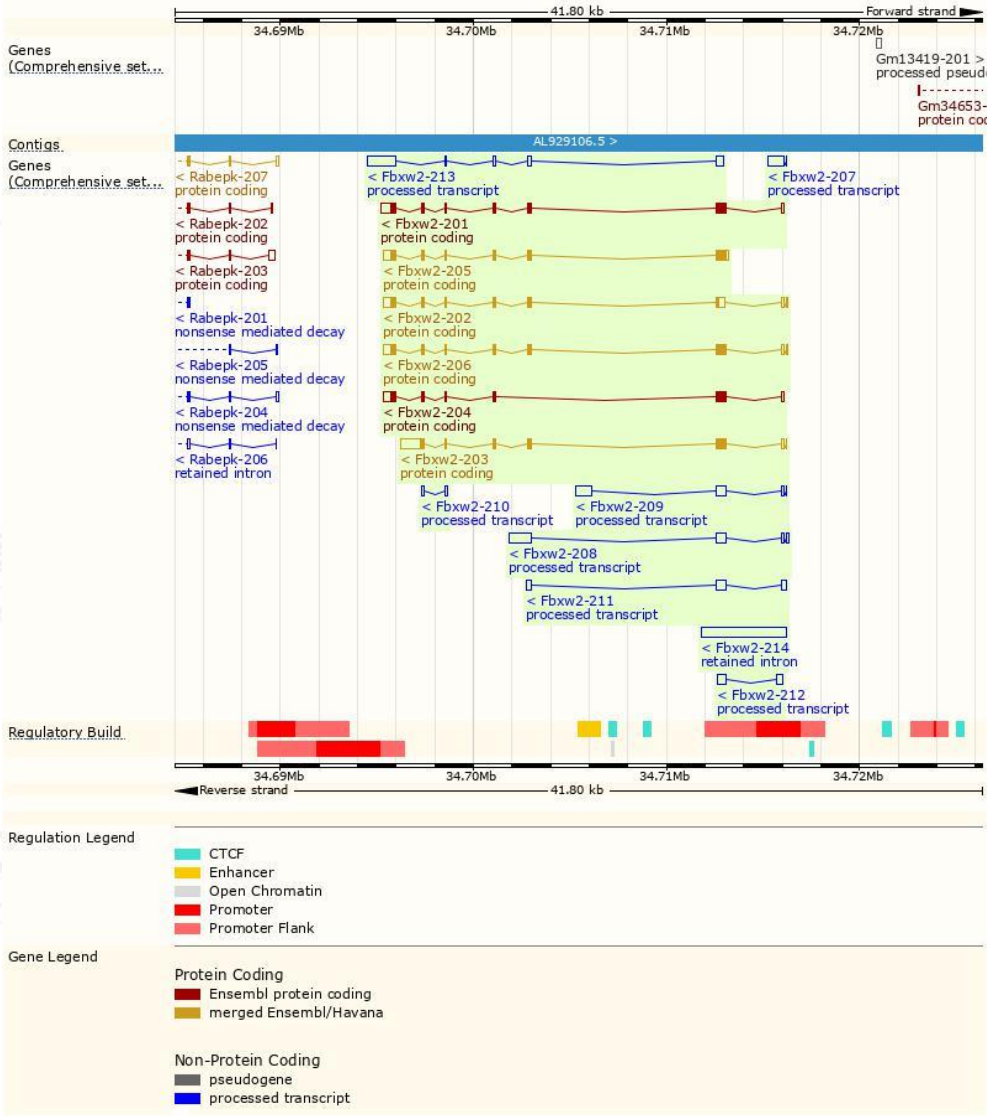
The gene has 14 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Fbxw2-203	ENSMUST00000113075.8	2260	359aa	Protein coding	CCDS50574		TSL:1 , GENCODE basic ,
Fbxw2-201	ENSMUST00000028220.10	2030	454aa	Protein coding	CCDS15952		TSL:1 , GENCODE basic , APPRIS P1 ,
Fbxw2-206	ENSMUST00000113080.9	1923	454aa	Protein coding	CCDS15952		TSL:1 , GENCODE basic , APPRIS P1 ,
Fbxw2-205	ENSMUST00000113078.8	1872	454aa	Protein coding	CCDS15952		TSL:1 , GENCODE basic , APPRIS P1 ,
Fbxw2-202	ENSMUST00000091020.10	1838	325aa	Protein coding	CCDS50573		TSL:1 , GENCODE basic ,
Fbxw2-204	ENSMUST00000113077.8	1670	389aa	Protein coding	-		TSL:5 , GENCODE basic ,
Fbxw2-213	ENSMUST00000156130.2	2265	No protein	Processed transcript	-		TSL:1 ,
Fbxw2-208	ENSMUST00000131534.8	1930	No protein	Processed transcript	-		TSL:1 ,
Fbxw2-209	ENSMUST00000134323.2	1521	No protein	Processed transcript	-		TSL:1 ,
Fbxw2-211	ENSMUST00000145660.8	988	No protein	Processed transcript	-		TSL:1 ,
Fbxw2-207	ENSMUST00000126051.2	852	No protein	Processed transcript	-		TSL:1 ,
Fbxw2-212	ENSMUST00000150865.2	807	No protein	Processed transcript	-		TSL:2 ,
Fbxw2-210	ENSMUST00000145180.2	320	No protein	Processed transcript	-		TSL:5 ,
Fbxw2-214	ENSMUST00000201806.2	4405	No protein	Retained intron	-		TSL:NA ,

The strategy is based on the design of *Fbxw2-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain

ENSMUSP00000028...

[Low complexity \(Seq\)](#)

[Superfamily](#)

[SMART](#)

[Prints](#)

[Pfam](#)

[PROSITE profiles](#)

[PROSITE patterns](#)

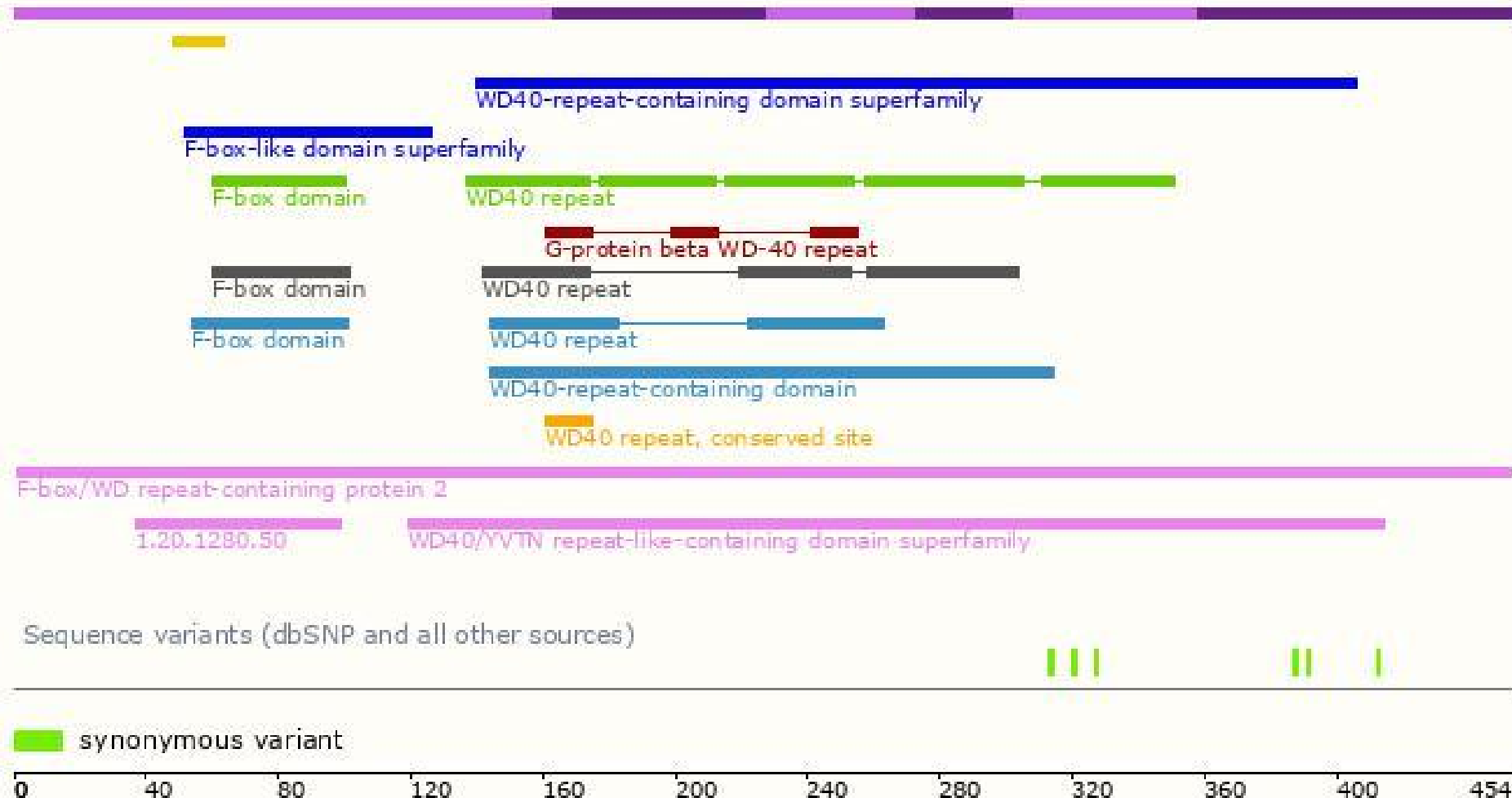
[PANTHER](#)

[Gene3D](#)

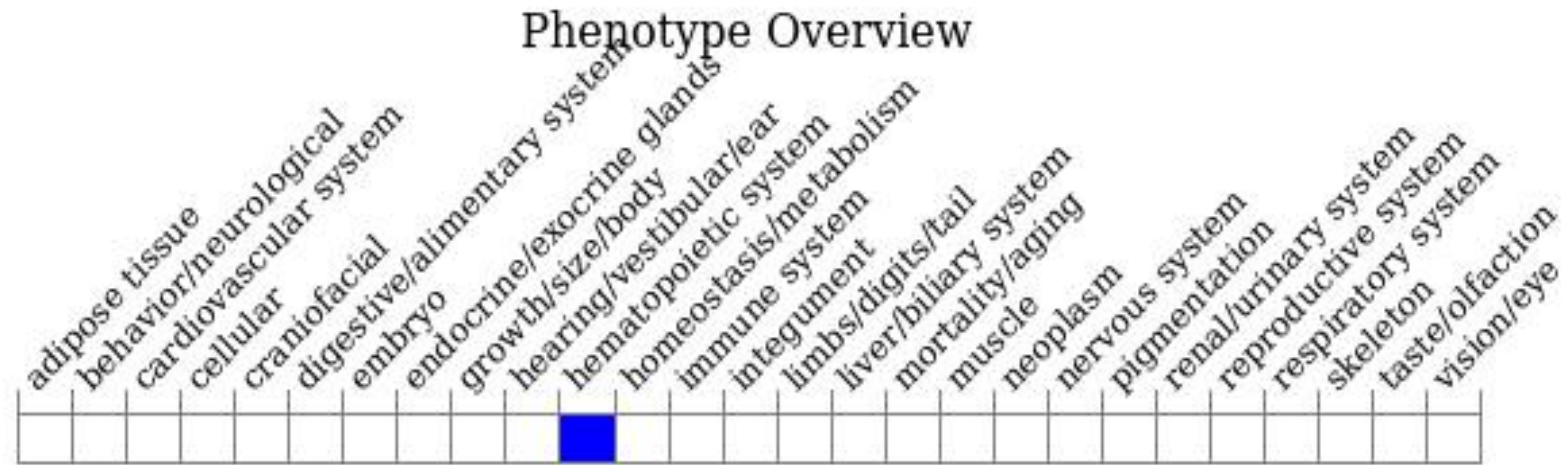
[All sequence SNPs/i...](#)

[Variant Legend](#)

[Scale bar](#)



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).

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

Tel: 025-5864 1534

