

# Fbxo22 Cas9-CKO Strategy

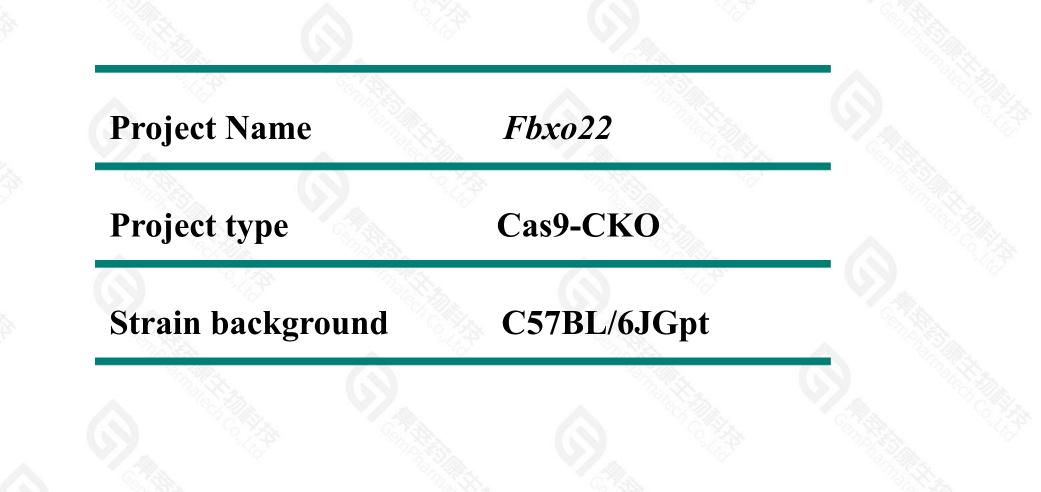
**Designer:Shuang Zhang** 

**Reviewer: Yun Li** 

**Design Date: 2020-5-24** 

## **Project Overview**



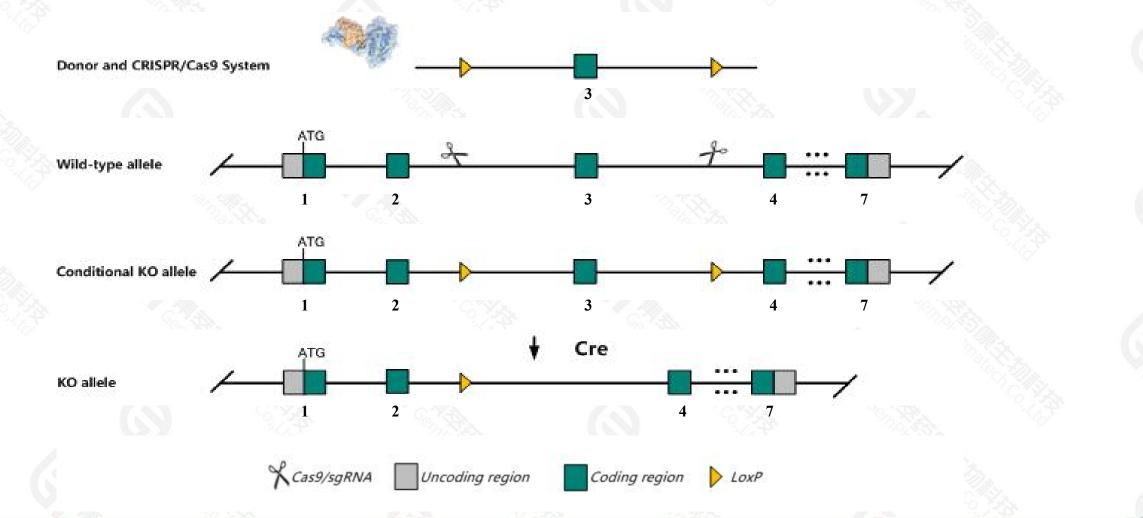


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## **Conditional Knockout strategy**

This model will use CRISPR/Cas9 technology to edit the Fbxo22 gene. The schematic diagram is as follows:



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### **Technical routes**



The *Fbxo22* gene has 5 transcripts. According to the structure of *Fbxo22* gene, exon3 of *Fbxo22*-201(ENSMUST00000034859.15) transcript is recommended as the knockout region. The region contains 88bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Fbxo22* 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 *Fbxo22* gene is located on the Chr9. 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)**



☆ ?

### Fbxo22 F-box protein 22 [Mus musculus (house mouse)]

Gene ID: 71999, updated on 3-Oct-2020

#### Summary

Official Symbol	Fbxo22 provided by MGI
<b>Official Full Name</b>	F-box protein 22 provided by <u>MGI</u>
Primary source	MGI:MGI:1926014
See related	Ensembl:ENSMUSG0000032309
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	0610033L19Rik, 1600016C16Rik
Expression	Ubiquitous expression in placenta adult (RPKM 12.5), CNS E11.5 (RPKM 7.6) and 28 other tissuesSee more
Orthologs	human all

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## **Transcript information (Ensembl)**

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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Fbxo22-201	ENSMUST0000034859.15	2112	<u>402aa</u>	Protein coding	CCD523202		TSL:1 , GENCODE basic , APPRIS P1 ,
Fbxo22-204	ENSMUST00000146201.8	2009	<u>299aa</u>	Protein coding	-		TSL:1 , GENCODE basic ,
Fbxo22-202	ENSMUST00000133795.2	476	<u>108aa</u>	Protein coding	1		CDS 3' incomplete , TSL:5 ,
Fbxo22-205	ENSMUST00000153970.8	1979	<u>41aa</u>	Nonsense mediated decay			TSL:1,
Fbxo22-203	ENSMUST00000140375.8	1921	<u>166aa</u>	Nonsense mediated decay	-		TSL:5 ,

The strategy is based on the design of *Fbxo22-201* transcript, the transcription is shown below:

			15.51 kb		- Forward strand
Fbxo22-201 > protein coding		1			
protein coung					

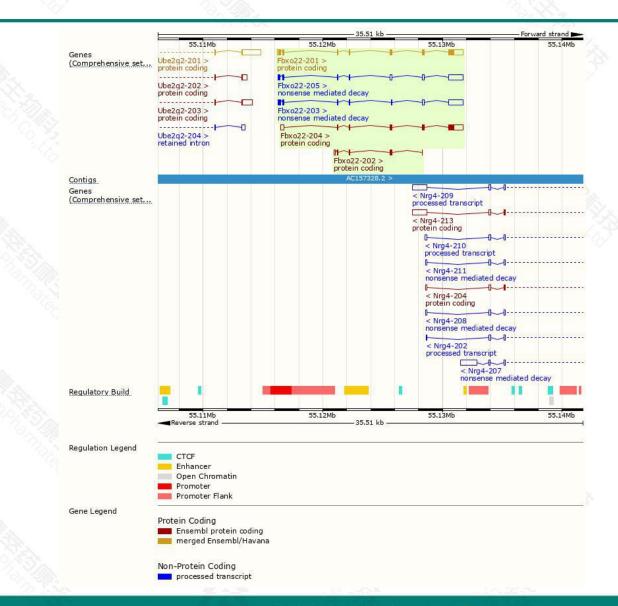
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### **Genomic location distribution**





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## **Protein domain**

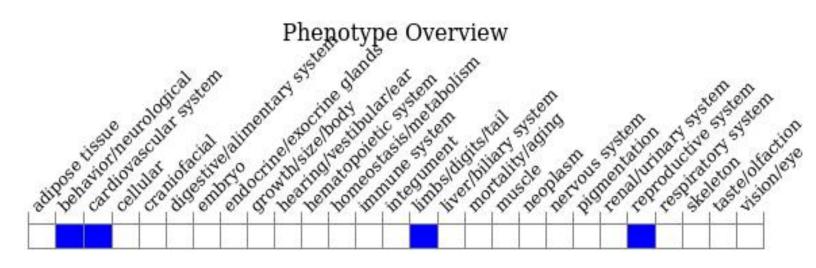


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	a <u>.</u> 12				8	FIST, C-domai	n Sil		34	
fam.	F-box domain	FIST, C-domain								
ANTHER	PTHR14939									-
ene3D	PTHR14939:SF5 1.20,1280,50									
ll sequence SNPs/i	Sequence variants (	dbSNP and a	ll other source	es)	10	101	÷1	TE		
/ariant Legend	missense varia splice region v synonymous v	ariant								
		68		55	200	112	100	320	395	

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### 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. Tel: 025-5864 1534



