

# Rnf123 Cas9-CKO Strategy

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**Reviewer: Xueting Zhang** 

**Design Date: 2020-11-6** 

## **Project Overview**



**Project Name** 

**Rnf123** 

**Project type** 

Cas9-CKO

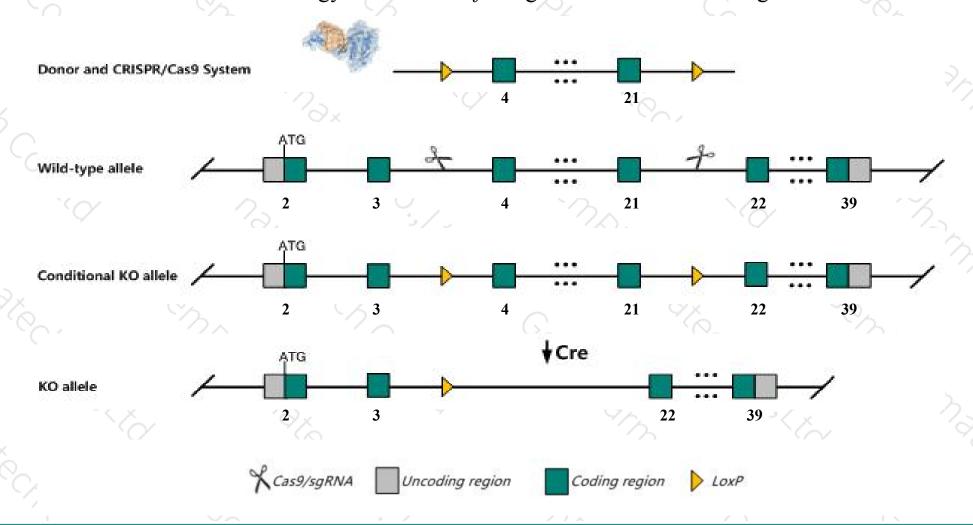
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rnf123* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Rnf123* gene has 18 transcripts. According to the structure of *Rnf123* gene, exon4-exon21 of *Rnf123*-214(ENSMUST00000162355.7) transcript is recommended as the knockout region. The region contains 1685bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Rnf123* 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**



- > Transcript Rnf123-204 CDS 3' incomplete and effect of Rnf123-204 is unknown.
- > The *Rnf123* 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.

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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)



#### Rnf123 ring finger protein 123 [ Mus musculus (house mouse) ]

Gene ID: 84585, updated on 26-Sep-2020

#### Summary

☆ ?

Official Symbol Rnf123 provided by MGI

Official Full Name ring finger protein 123 provided by MGI

Primary source MGI:MGI:2148796

See related Ensembl: ENSMUSG00000041528

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as KPC; Kpc1; BC003945

Expression Ubiquitous expression in liver E14.5 (RPKM 28.3), spleen adult (RPKM 26.5) and 28 other tissues See more

Orthologs human all

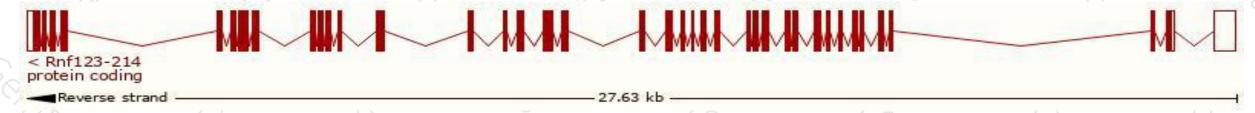
## Transcript information (Ensembl)



#### The gene has 18 transcripts, all transcripts are shown below:

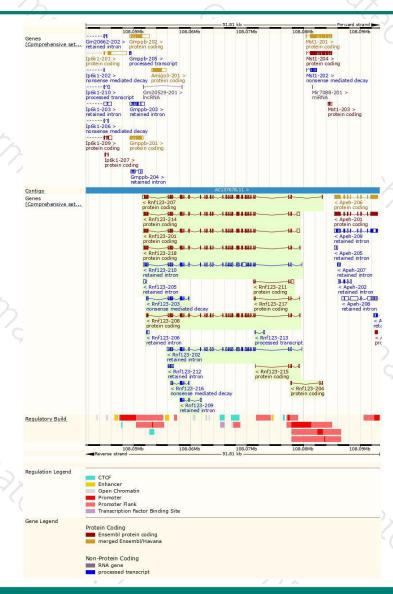
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rnf123-214	ENSMUST00000162355.7	4727	1320aa	Protein coding	CCD581072	Q5XPI3	TSL:1 GENCODE basic APPRIS ALT2
Rnf123-207	ENSMUST00000160249.7	4426	1314aa	Protein coding	CCDS52922	Q5XPI3	TSL:5 GENCODE basic APPRIS P3
Rnf123-218	ENSMUST00000178267.7	4309	1314aa	Protein coding	CCDS52922	Q5XPI3	TSL:5 GENCODE basic APPRIS P3
Rnf123-201	ENSMUST00000047746.12	4722	1320aa	Protein coding	-	Q5XPI3	TSL:5 GENCODE basic APPRIS ALT2
Rnf123-208	ENSMUST00000160649.7	3919	1239aa	Protein coding	21	Q05CH9	CDS 3' incomplete TSL:1
Rnf123-211	ENSMUST00000161828.7	799	<u>111aa</u>	Protein coding	-	E0CXL3	CDS 3' incomplete TSL:2
Rnf123-204	ENSMUST00000159372.2	369	27aa	Protein coding	-	E0CYS5	CDS 3' incomplete TSL:2
Rnf123-215	ENSMUST00000162516.2	365	59aa	Protein coding	-	E0CX26	CDS 3' incomplete TSL:5
Rnf123-217	ENSMUST00000174504.7	347	<u>91aa</u>	Protein coding	-	G3UYH5	CDS 3' incomplete TSL:5
Rnf123-203	ENSMUST00000159306.7	1137	<u>261aa</u>	Nonsense mediated decay		F7B5Z3	CDS 5' incomplete TSL:1
Rnf123-216	ENSMUST00000162753.7	719	61aa	Nonsense mediated decay	2	F6U715	CDS 5' incomplete TSL:3
Rnf123-213	ENSMUST00000162152.1	332	No protein	Processed transcript	-	1-1	TSL:5
Rnf123-210	ENSMUST00000161673.7	4658	No protein	Retained intron	21	725	TSL:2
Rnf123-202	ENSMUST00000159136.7	3789	No protein	Retained intron	-	(7)	TSL:1
Rnf123-209	ENSMUST00000160841.1	745	No protein	Retained intron		-	TSL:2
Rnf123-212	ENSMUST00000162123.1	545	No protein	Retained intron	-	22	TSL:2
Rnf123-205	ENSMUST00000159523.1	445	No protein	Retained intron	-	(70)	TSL:2
Rnf123-206	ENSMUST00000159526.1	322	No protein	Retained intron	-8	-	TSL:2

The strategy is based on the design of *Rnf123-214* transcript, the transcription is shown below:



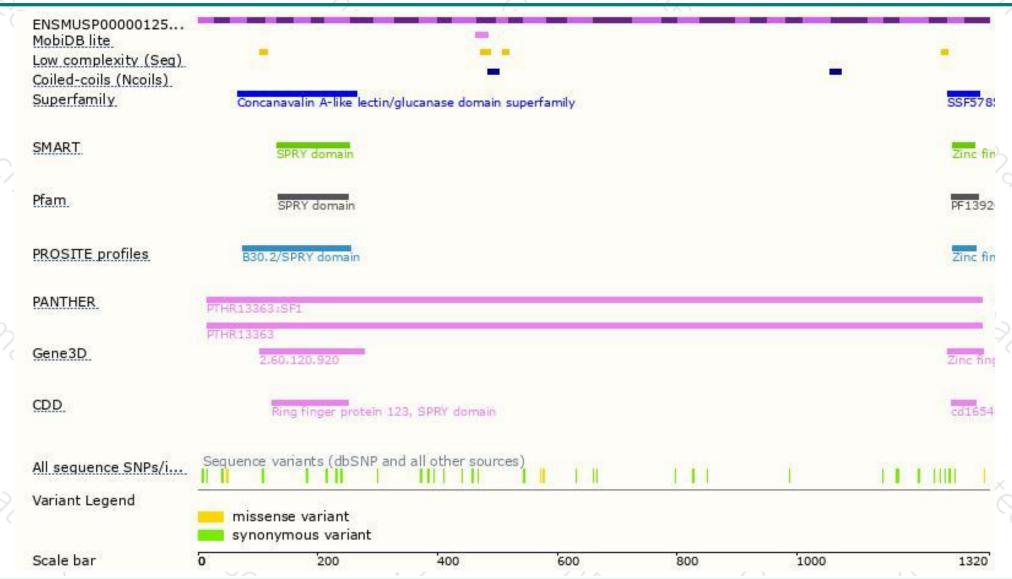
## Genomic location distribution





### Protein domain







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

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