

***Rnf44* Cas9-CKO Strategy**

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Design Date: 2020-9-16

Project Overview

Project Name

Rnf44

Project type

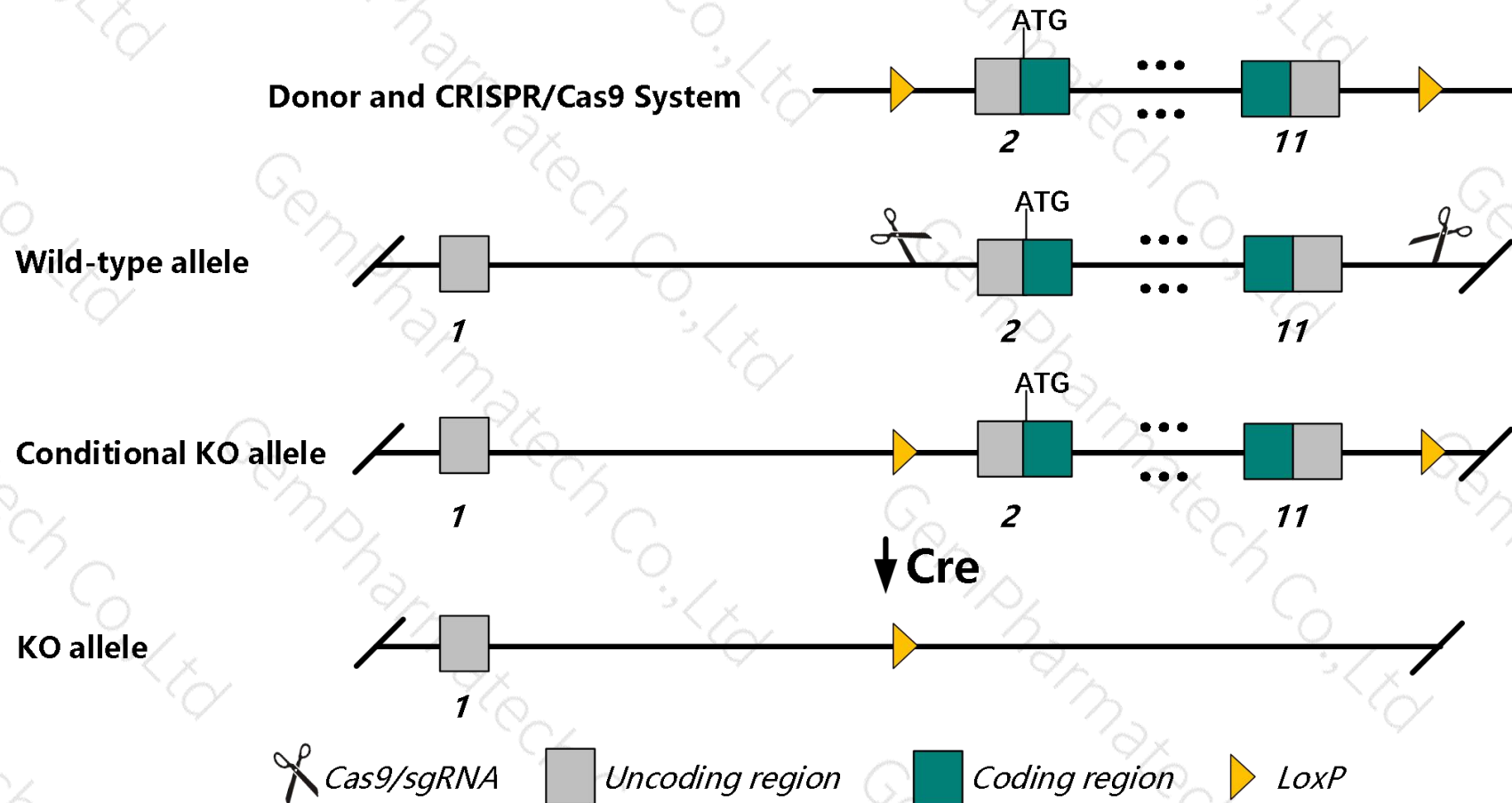
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rnf44* gene. The schematic diagram is as follows:



Technical routes

- The *Rnf44* gene has 18 transcripts. According to the structure of *Rnf44* gene, exon2-exon11 of *Rnf44*-204(ENSMUST00000125871.7) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rnf44* 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 *Rnf44* gene is located on the Chr13. 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)

Rnf44 ring finger protein 44 [Mus musculus (house mouse)]

Gene ID: 105239, updated on 13-Mar-2020

Summary



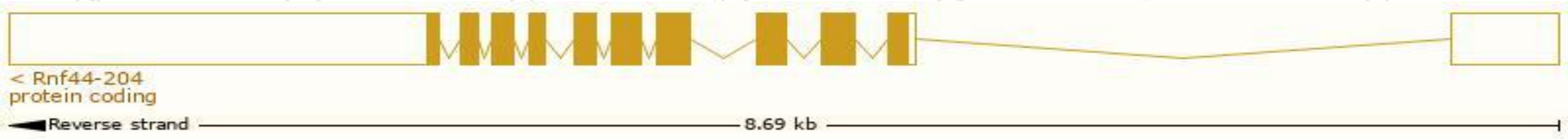
Official Symbol	Rnf44 provided by MGI
Official Full Name	ring finger protein 44 provided by MGI
Primary source	MGI:MGI:2145310
See related	Ensembl:ENSMUSG00000034928
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI854545, mKIAA1100
Expression	Ubiquitous expression in adrenal adult (RPKM 91.4), thymus adult (RPKM 80.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

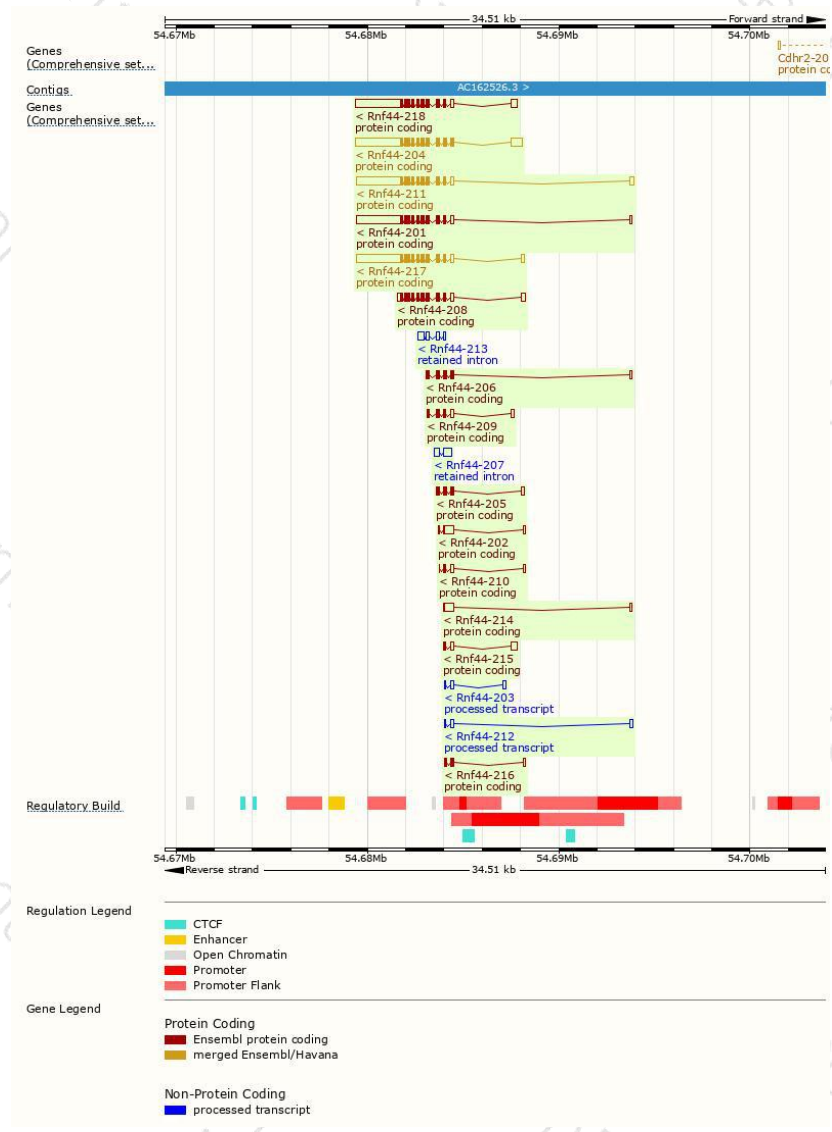
The gene has 18 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rnf44-204	ENSMUST00000125871.7	4292	432aa	Protein coding	CCDS49268	E9PUC0	TSL:1 GENCODE basic APPRIS P4
Rnf44-218	ENSMUST00000177950.7	3941	350aa	Protein coding	CCDS49266	Q3UJH8	TSL:1 GENCODE basic APPRIS ALT1
Rnf44-211	ENSMUST00000134862.7	3797	351aa	Protein coding	CCDS49267	Q3UJH8	TSL:5 GENCODE basic APPRIS ALT1
Rnf44-217	ENSMUST00000150806.7	3772	351aa	Protein coding	CCDS49267	Q3UJH8	TSL:1 GENCODE basic APPRIS ALT1
Rnf44-201	ENSMUST00000037422.15	3734	350aa	Protein coding	CCDS49266	Q3UJH8	TSL:1 GENCODE basic APPRIS ALT1
Rnf44-208	ENSMUST00000128257.7	1689	350aa	Protein coding	CCDS49266	Q3UJH8	TSL:5 GENCODE basic APPRIS ALT1
Rnf44-206	ENSMUST00000126717.7	792	210aa	Protein coding	-	D3Z721	CDS 3' incomplete TSL:3
Rnf44-209	ENSMUST00000129881.7	698	103aa	Protein coding	-	D3Z7N0	CDS 3' incomplete TSL:3
Rnf44-202	ENSMUST00000122935.1	671	37aa	Protein coding	-	D3Z6D0	CDS 3' incomplete TSL:3
Rnf44-205	ENSMUST00000125927.7	664	150aa	Protein coding	-	D3Z6V1	CDS 3' incomplete TSL:3
Rnf44-214	ENSMUST00000145397.1	609	17aa	Protein coding	-	D3YYQ1	CDS 3' incomplete TSL:2
Rnf44-215	ENSMUST00000146931.7	550	7aa	Protein coding	-	A0A1Y7VGK4	CDS 3' incomplete TSL:2
Rnf44-210	ENSMUST00000134177.7	386	19aa	Protein coding	-	D3YXW8	CDS 3' incomplete TSL:5
Rnf44-216	ENSMUST00000150626.1	354	69aa	Protein coding	-	D3Z261	CDS 3' incomplete TSL:3
Rnf44-203	ENSMUST00000123333.7	373	No protein	Processed transcript	-	-	TSL:2
Rnf44-212	ENSMUST00000144052.1	322	No protein	Processed transcript	-	-	TSL:3
Rnf44-213	ENSMUST00000145215.1	758	No protein	Retained intron	-	-	TSL:3
Rnf44-207	ENSMUST00000128038.1	725	No protein	Retained intron	-	-	TSL:2

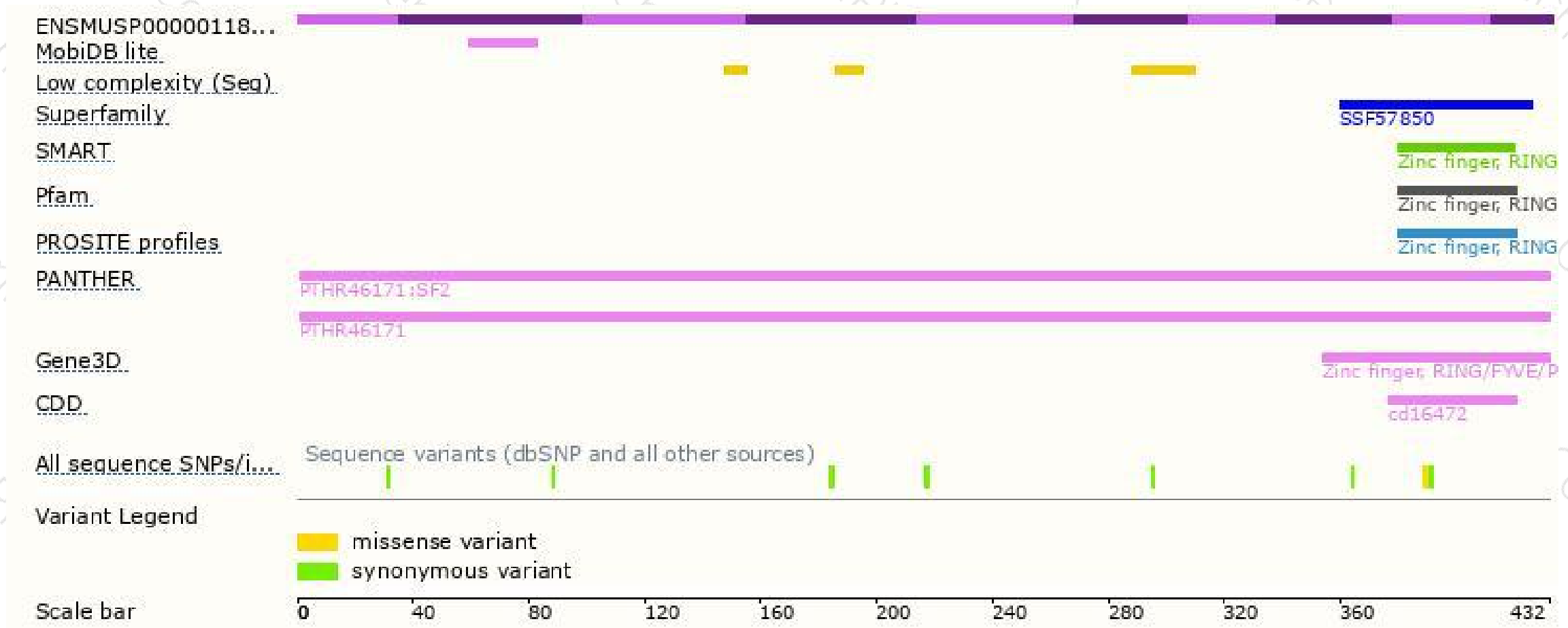
The strategy is based on the design of *Rnf44-204* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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