

***Zfp236* Cas9-CKO Strategy**

Designer: Jia Yu

Reviewer: Xiaojing Li

Design Date: 2020-10-13

Project Overview

Project Name

Zfp236

Project type

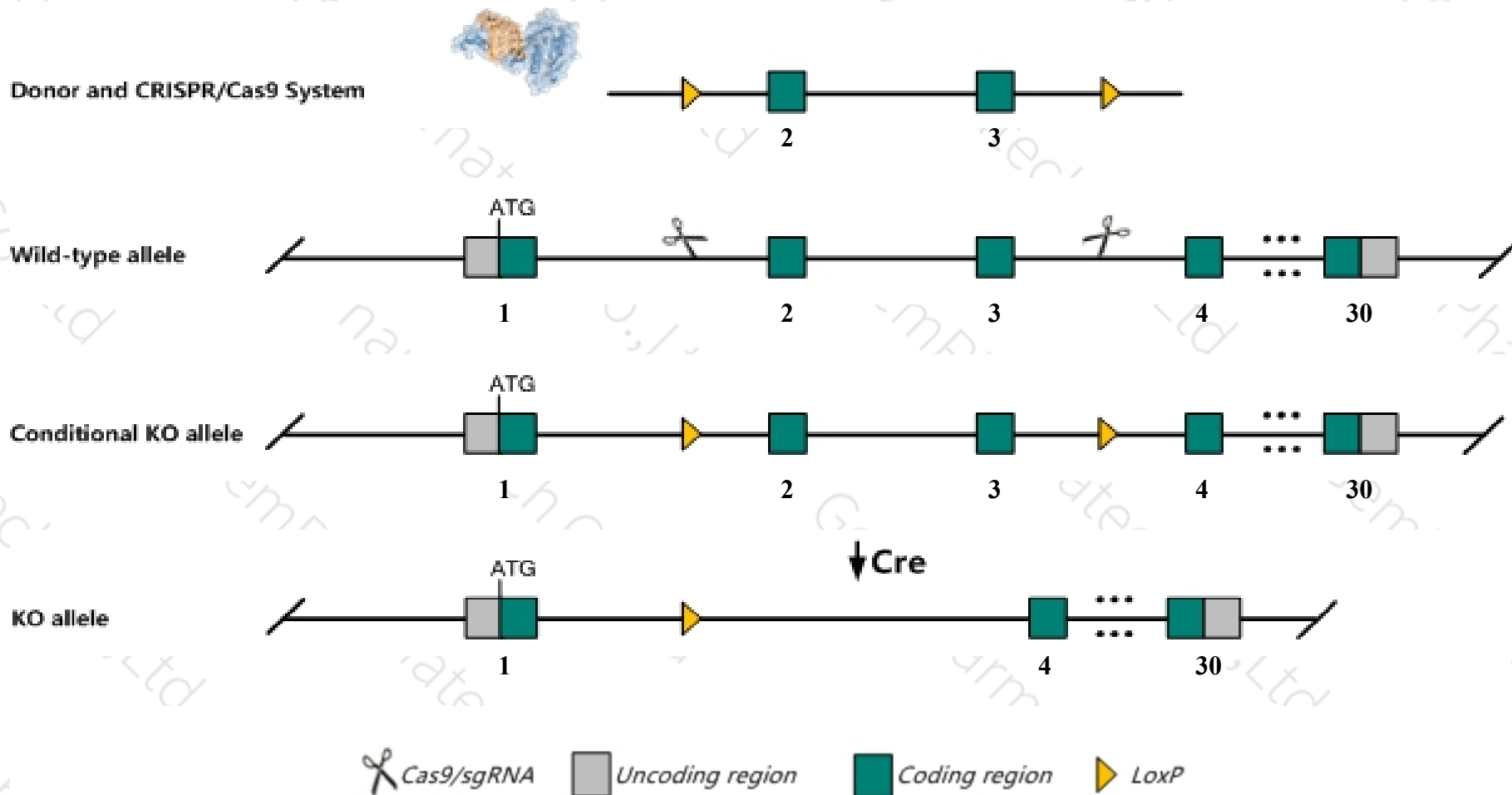
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Zfp236* gene. The schematic diagram is as follows:



Technical routes

- The *Zfp236* gene has 7 transcripts. According to the structure of *Zfp236* gene, exon2-exon3 of *Zfp236-201*(ENSMUST00000171071.8) transcript is recommended as the knockout region. The region contains 308bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Zfp236* 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.

- The *Zfp236* gene is located on the Chr18. 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)

Zfp236 zinc finger protein 236 [Mus musculus (house mouse)]

Gene ID: 329002, updated on 13-Mar-2020

Summary



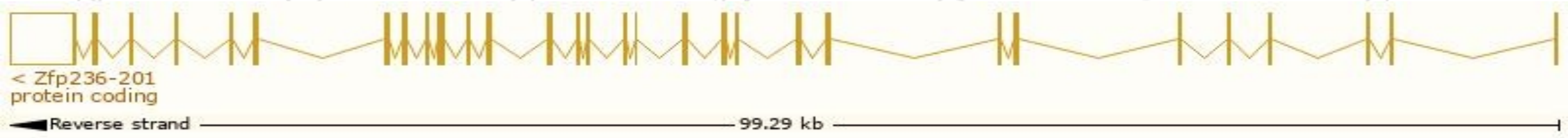
Official Symbol	Zfp236 provided by MGI
Official Full Name	zinc finger protein 236 provided by MGI
Primary source	MGI:MGI:1926950
See related	Ensembl:ENSMUSG00000041258
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI447957, Gm335
Expression	Ubiquitous expression in thymus adult (RPKM 8.4), ovary adult (RPKM 4.7) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

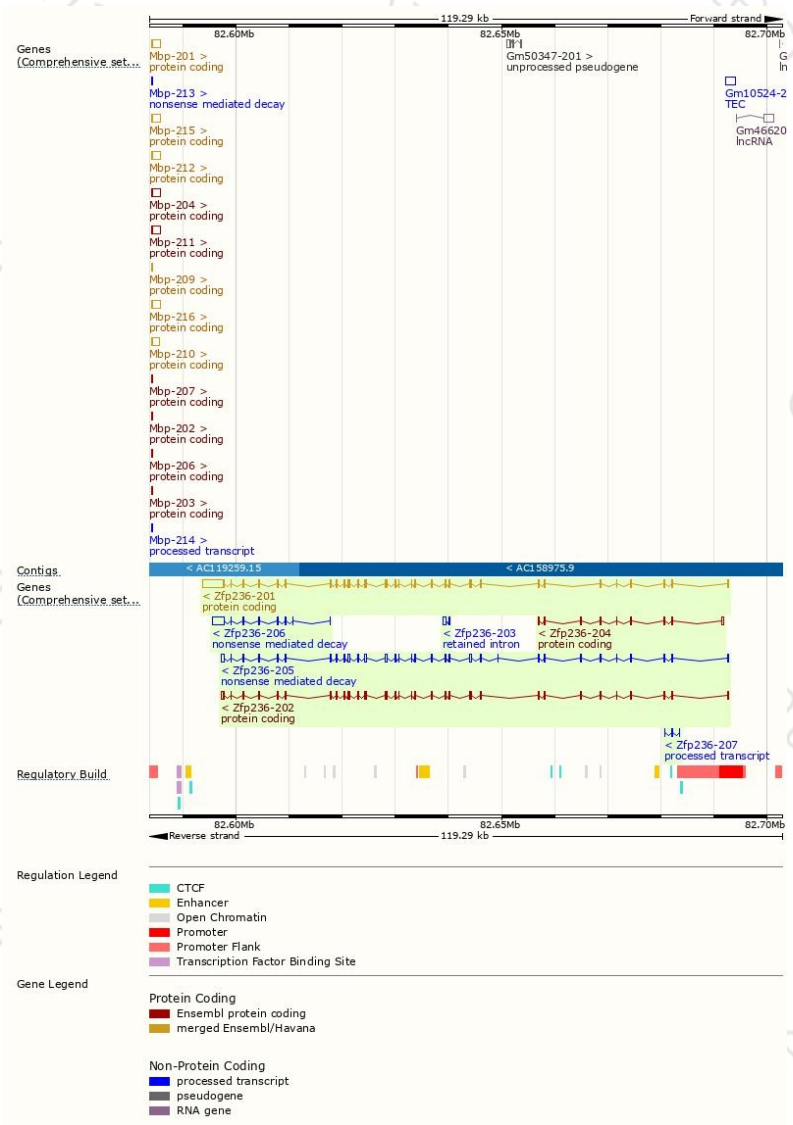
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Zfp236-201	ENSMUST00000171071.8	9626	1799aa	Protein coding	CCDS50337	B2RR24	TSL:1 GENCODE basic APPRIS P2
Zfp236-202	ENSMUST00000182122.7	6059	1847aa	Protein coding	-	S4R299	TSL:1 GENCODE basic APPRIS ALT2
Zfp236-204	ENSMUST00000182866.2	1503	429aa	Protein coding	-	S4R1R9	CDS 3' incomplete TSL:3
Zfp236-205	ENSMUST00000183048.7	6170	485aa	Nonsense mediated decay	-	S4R1D8	TSL:1
Zfp236-206	ENSMUST00000183324.7	3227	43aa	Nonsense mediated decay	-	S4R1Z3	CDS 5' incomplete TSL:1
Zfp236-207	ENSMUST00000225002.1	156	No protein	Processed transcript	-	-	
Zfp236-203	ENSMUST00000182759.1	665	No protein	Retained intron	-	-	TSL:5

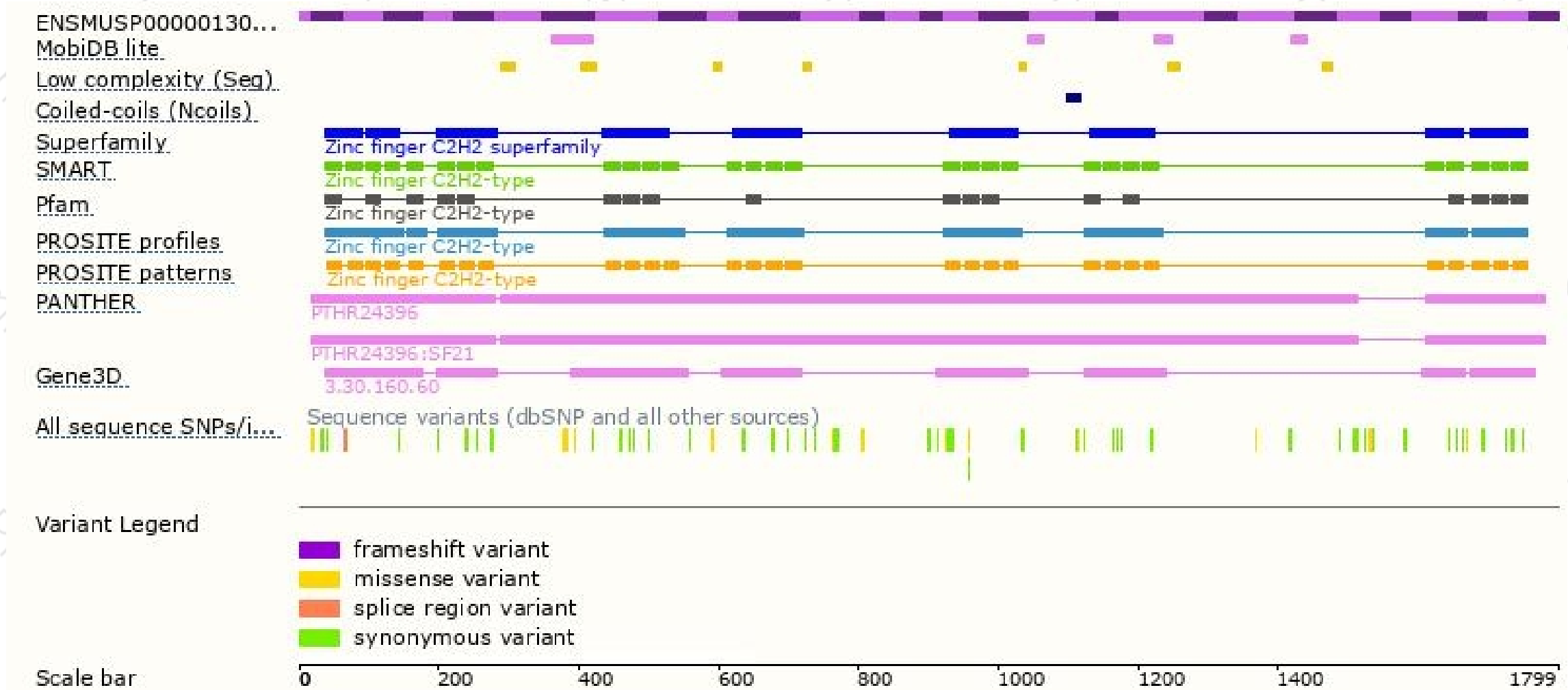
The strategy is based on the design of *Zfp236-201* transcript,the transcription is shown below:



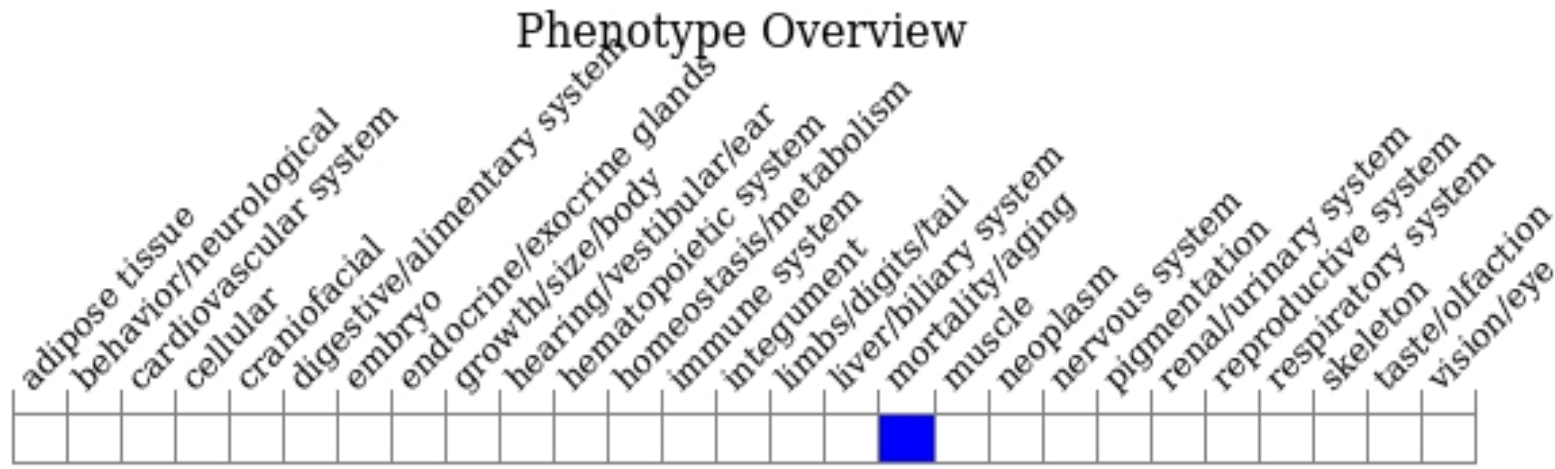
Genomic location distribution



Protein domain



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

