Zwilch Cas9-CKO Strategy

Designer: Daohua Xu
Reviewer: Huimin Su

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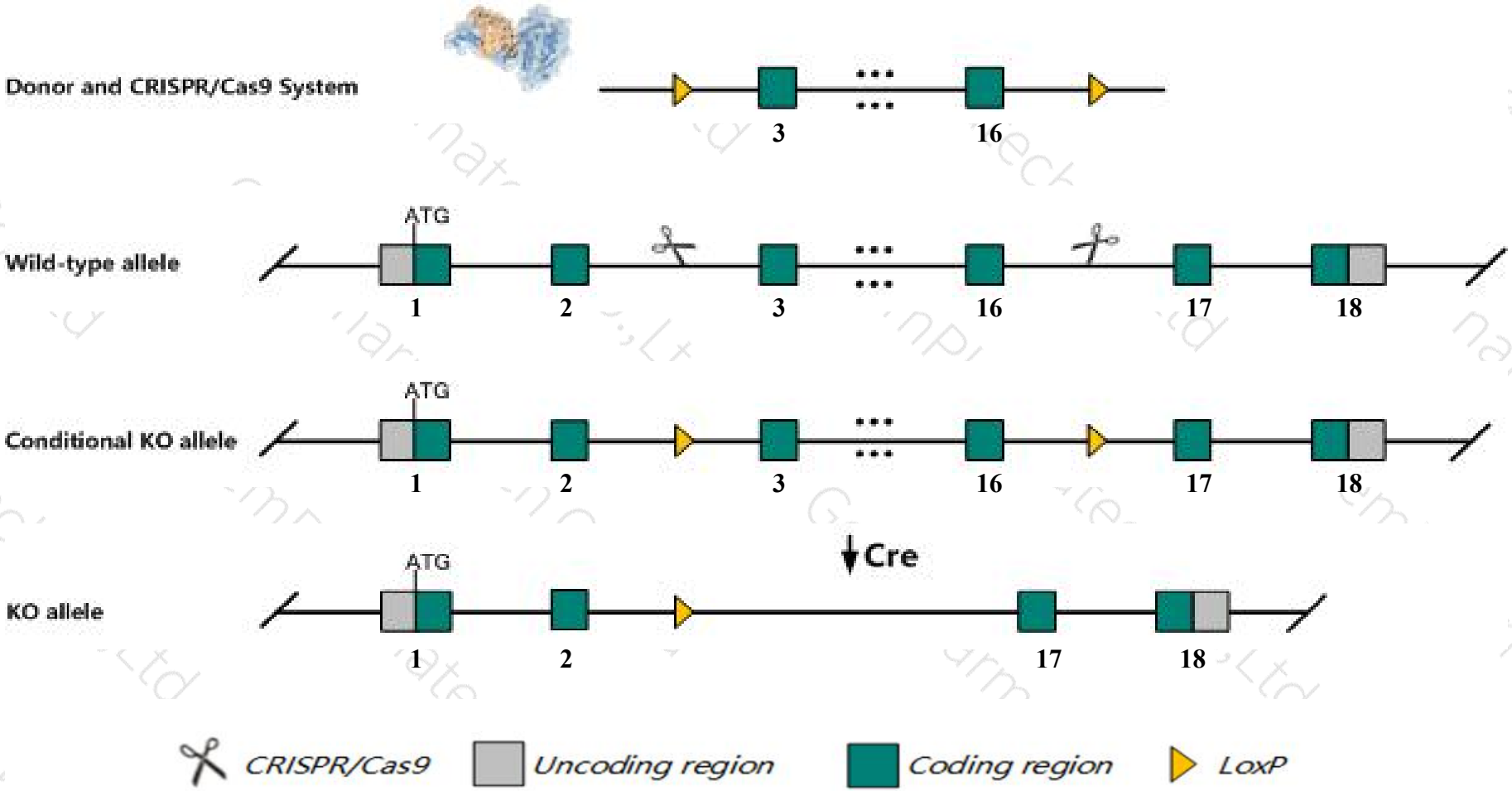
Project Overview

Project Name	***Zwilch***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

- The *Zwilch* gene has 13 transcripts. According to the structure of *Zwilch* gene, exon3-exon16 of *Zwilch-201* (ENSMUST00000122091.7) transcript is recommended as the knockout region.The region contains 1463bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Zwilch* gene. The brief process is as follows:CRISPR/Cas9 system 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 will be 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 *Zwilch* 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）

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Zwilch zwilch kinetochore protein [Mus musculus (house mouse)]

Gene ID: 68014, updated on 31-Jan-2019

Summary

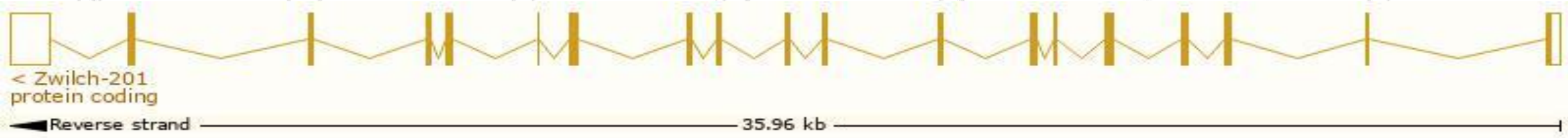
Official Symbol	Zwilch provided by MGI
Official Full Name	zwilch kinetochore protein provided by MGI
Primary source	MGI:MGI:1915264
See related	Ensembl:ENSMUSG00000032400
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2310031L18Rik
Expression	Broad expression in liver E14 (RPKM 15.3), CNS E11.5 (RPKM 11.6) and 15 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

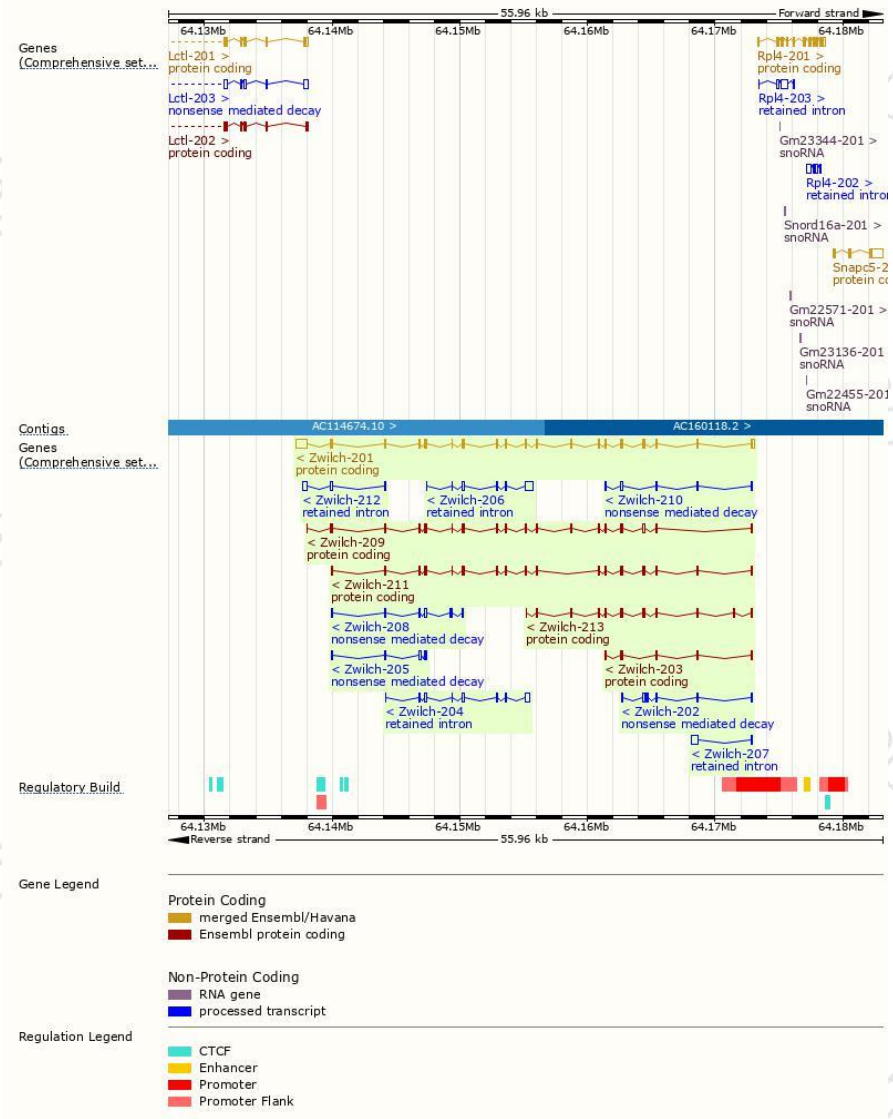
The gene has 13 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Zwilch-201	ENSMUST00000122091.7	2957	596aa	Protein coding	CCDS23275	G3X9Z0	TSL:1 GENCODE basic APPRIS P2
Zwilch-209	ENSMUST00000176299.7	1831	475aa	Protein coding	-	H3BKZ0	TSL:5 GENCODE basic APPRIS ALT2
Zwilch-211	ENSMUST00000176794.7	1746	572aa	Protein coding	-	H3BJ52	TSL:5 GENCODE basic APPRIS ALT2
Zwilch-213	ENSMUST00000177045.7	1047	303aa	Protein coding	-	H3BKB9	CDS 3' incomplete TSL:1
Zwilch-203	ENSMUST00000130127.7	463	154aa	Protein coding	-	D3Z6K4	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Zwilch-208	ENSMUST00000152824.7	593	51aa	Nonsense mediated decay	-	F6TCM4	CDS 5' incomplete TSL:3
Zwilch-202	ENSMUST00000124301.2	545	82aa	Nonsense mediated decay	-	H3BJF6	CDS 5' incomplete TSL:5
Zwilch-210	ENSMUST00000176378.8	508	86aa	Nonsense mediated decay	-	H3BIZ1	TSL:5
Zwilch-205	ENSMUST00000143421.1	368	38aa	Nonsense mediated decay	-	H3BL96	CDS 5' incomplete TSL:3
Zwilch-204	ENSMUST00000138075.8	944	No protein	Retained intron	-	-	TSL:5
Zwilch-206	ENSMUST00000148428.1	942	No protein	Retained intron	-	-	TSL:5
Zwilch-207	ENSMUST00000151428.1	638	No protein	Retained intron	-	-	TSL:2
Zwilch-212	ENSMUST00000176848.1	609	No protein	Retained intron	-	-	TSL:2

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

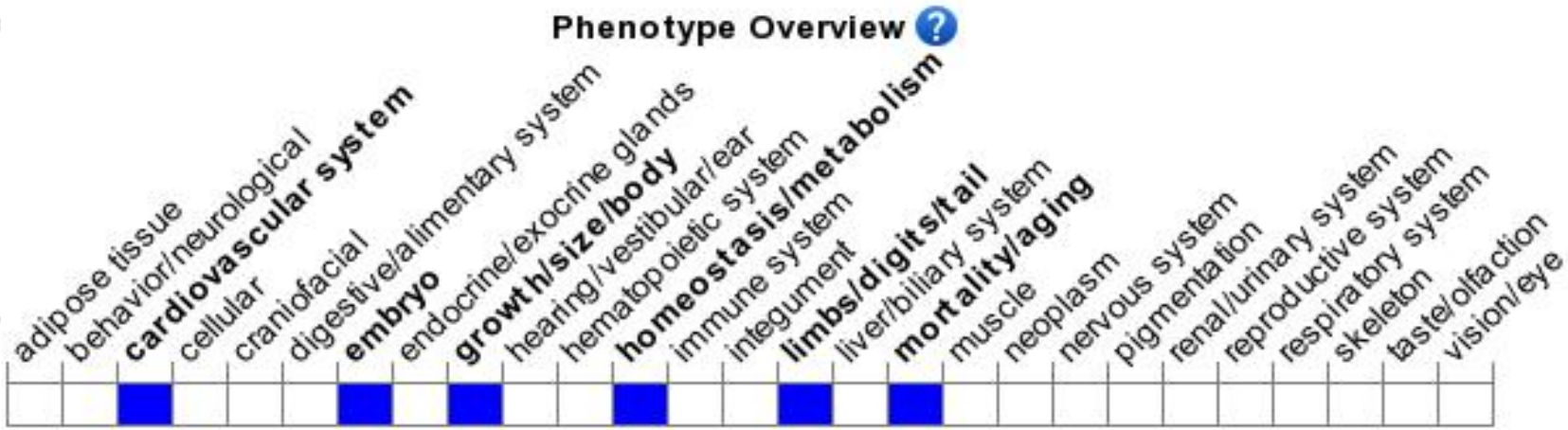


Genomic location distribution



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/).

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

Tel: 400-9660890

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GemPharmatech Co.,Ltd