Zeb1 Cas9-CKO Strategy

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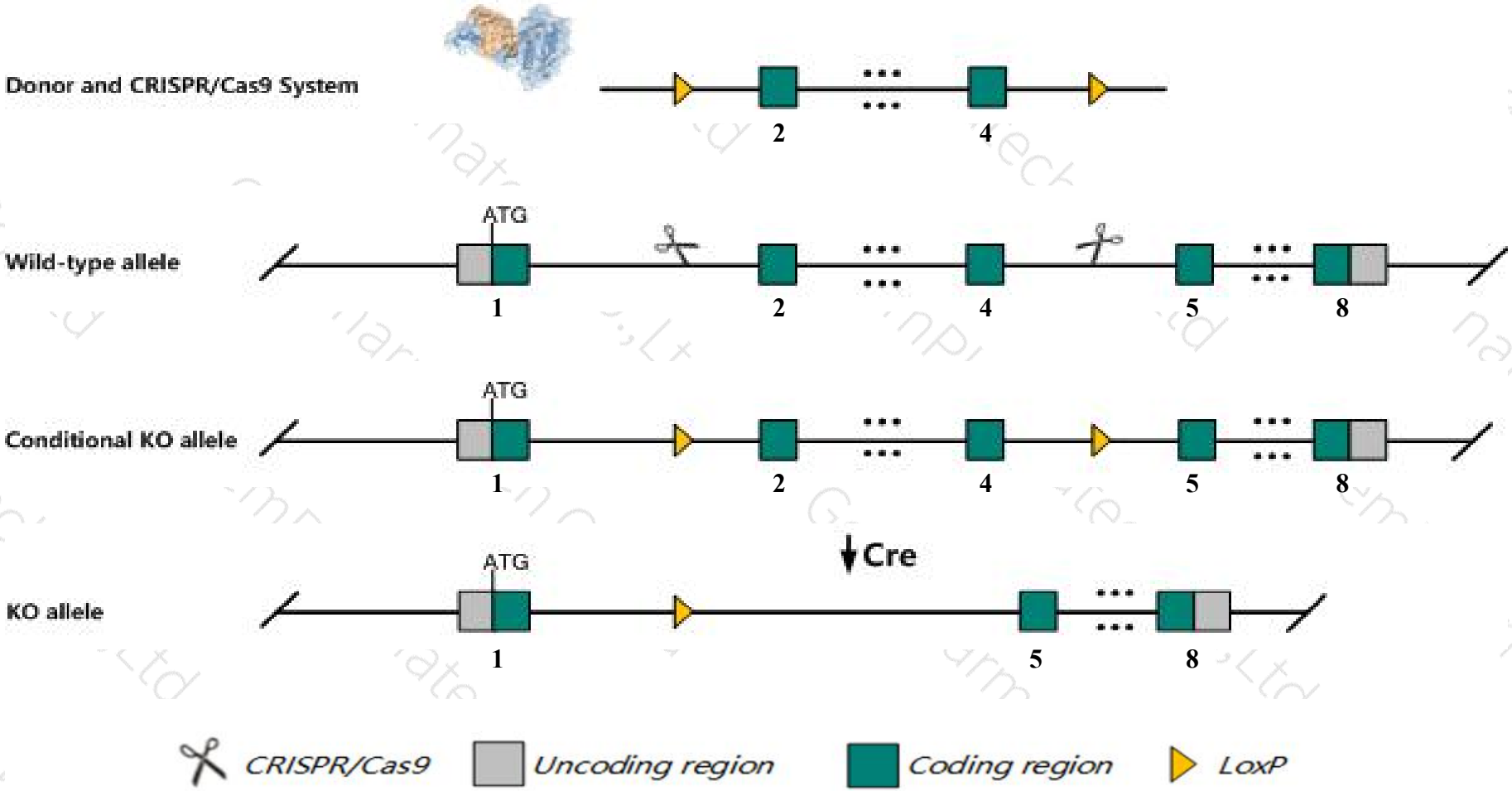
Project Overview

Project Name	***Zeb1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Zeb1* gene. The schematic diagram is as follows:



Technical routes

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- The *Zeb1* gene has 12 transcripts. According to the structure of *Zeb1* gene, exon2-exon4 of *Zeb1-201* (ENSMUST00000025081.12) transcript is recommended as the knockout region.The region contains 566bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Zeb1* 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

- According to the existing MGI data,Mutations at this locus affect thymus organization and homozygotes exhibit severe thymic T cell deficiency. Some mutations result in eye anomalies and extensive skeletal abnormalities. Homozygotes generally die at birth due to respiratory failure.

- The *Zeb1* 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）

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Zeb1 zinc finger E-box binding homeobox 1 [Mus musculus (house mouse)]

Gene ID: 21417, updated on 9-Apr-2019

Summary

Official Symbol	Zeb1 provided by MGI
Official Full Name	zinc finger E-box binding homeobox 1 provided by MGI
Primary source	MGI:MGI:1344313
See related	Ensembl:ENSMUSG00000024238
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	3110032K11Rik, AREB6, BZP, MEB1, Nil2, TCF-8, Tcf18, Tcf8, Tw, ZEB, Zfhep, Zfhx1a, Zfx1a, Zfx1ha, [delta]EF1
Expression	Ubiquitous expression in CNS E11.5 (RPKM 9.2), limb E14.5 (RPKM 8.6) and 26 other tissues See more
Orthologs	human all

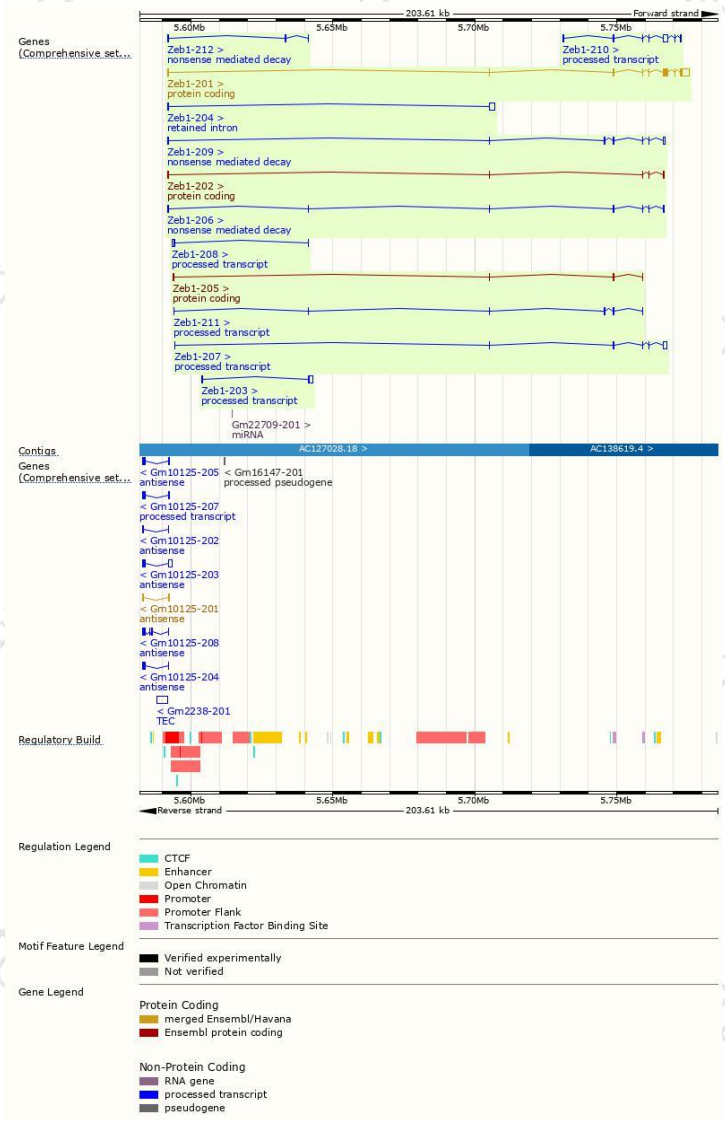
Transcript information（Ensembl）

The gene has 12 transcripts,all transcripts are shown below:

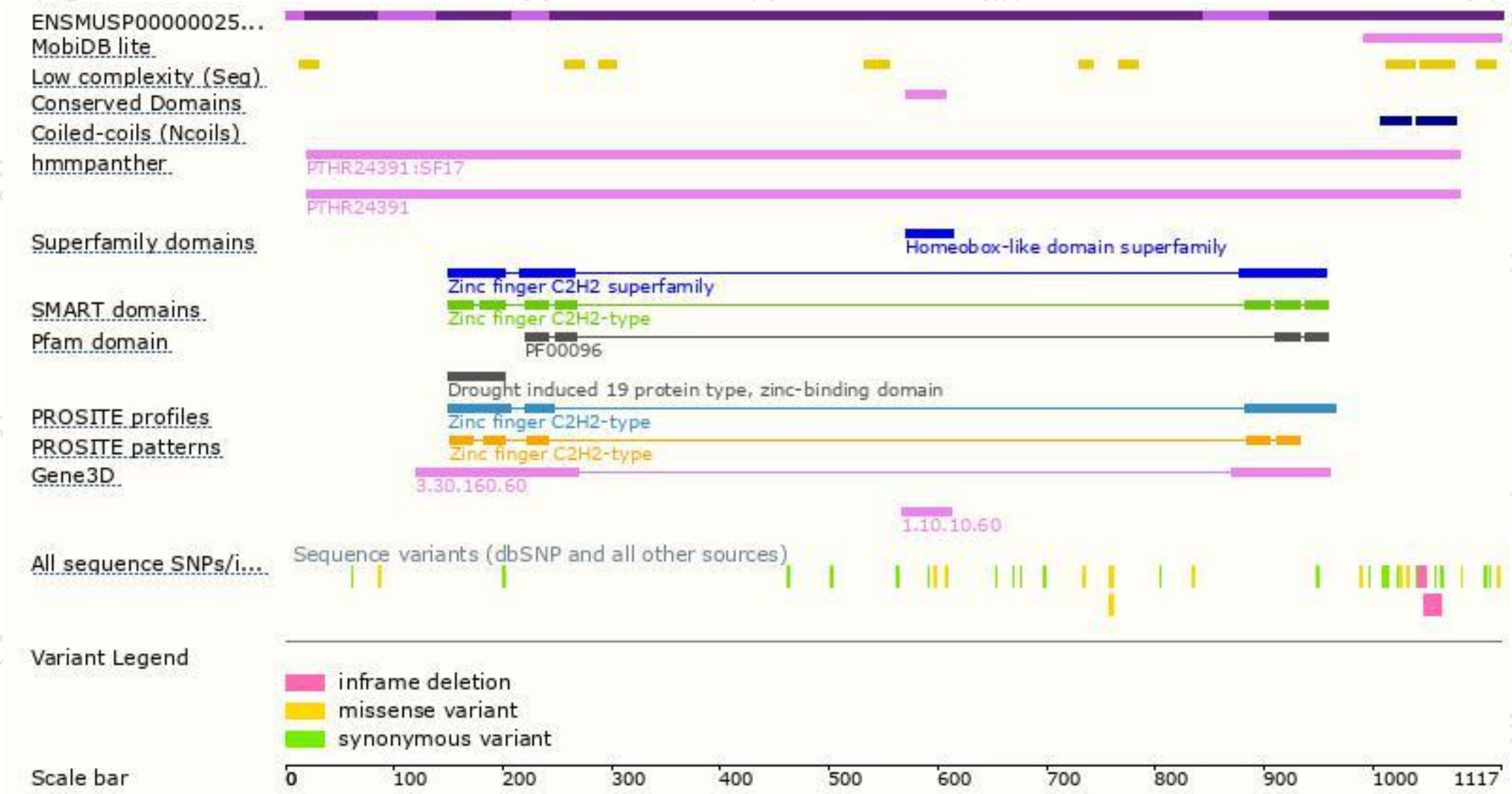
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Zeb1-201	ENSMUST00000025081.12	5801	1117aa	Protein coding	CCDS29039	Q64318	TSL:1 GENCODE basic APPRIS P1
Zeb1-202	ENSMUST00000159390.7	900	293aa	Protein coding	-	E9PXY5	CDS 3' incomplete TSL:3
Zeb1-205	ENSMUST00000160910.1	774	154aa	Protein coding	-	E0CX96	CDS 3' incomplete TSL:3
Zeb1-209	ENSMUST00000175925.7	1485	105aa	Nonsense mediated decay	-	H3BJU2	TSL:5
Zeb1-206	ENSMUST00000161295.7	1215	20aa	Nonsense mediated decay	-	E0CY37	CDS 5' incomplete TSL:5
Zeb1-212	ENSMUST00000224200.2	266	37aa	Nonsense mediated decay	-	-	
Zeb1-210	ENSMUST00000177030.1	2768	No protein	Processed transcript	-	-	TSL:5
Zeb1-207	ENSMUST00000162892.7	2464	No protein	Processed transcript	-	-	TSL:1
Zeb1-203	ENSMUST00000159477.1	1610	No protein	Processed transcript	-	-	TSL:1
Zeb1-211	ENSMUST00000177070.7	819	No protein	Processed transcript	-	-	TSL:5
Zeb1-208	ENSMUST00000175739.1	790	No protein	Processed transcript	-	-	TSL:3
Zeb1-204	ENSMUST00000160522.1	1938	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Zeb1-201* transcript,The transcription is shown below

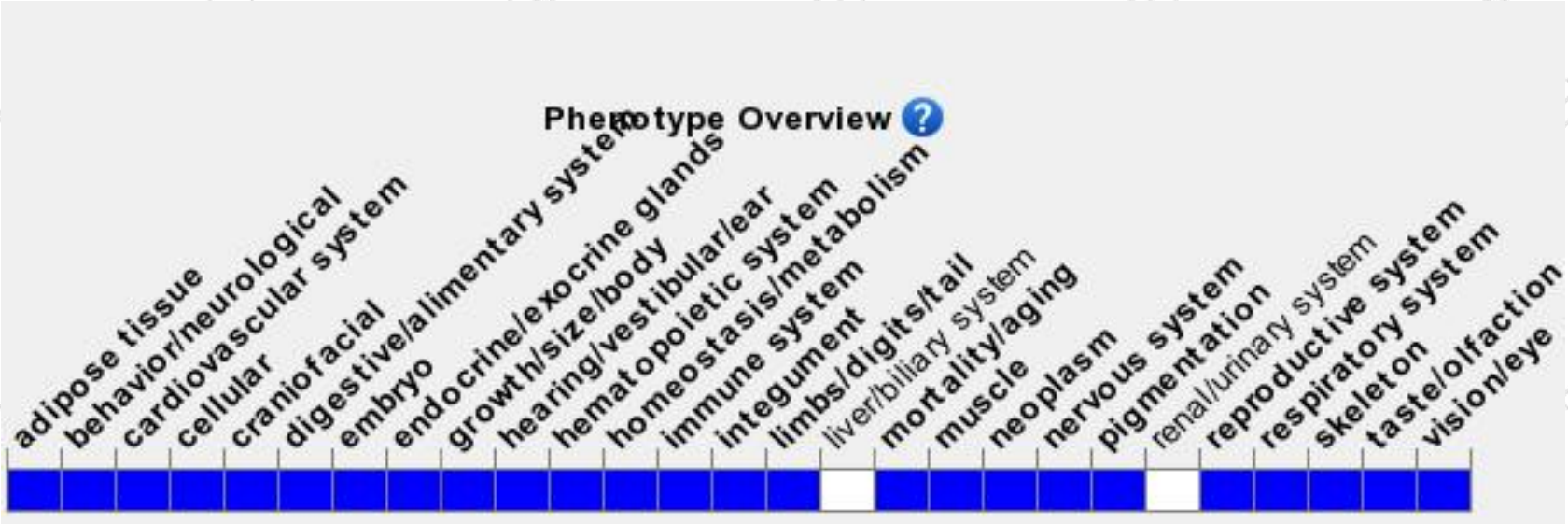
Genomic location distribution



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.

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