Zeb2 Cas9-KO Strategy

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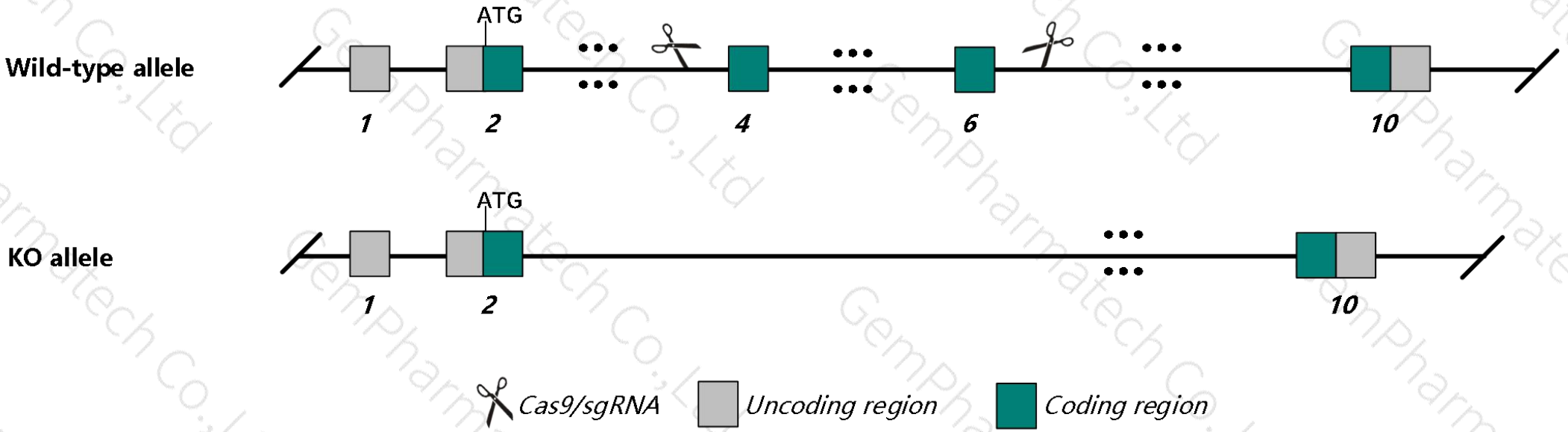
Design Date: 2019-11-14

Project Overview

Project Name	***Zeb2***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Zeb2* gene. The schematic diagram is as follows:



Technical routes

- The *Zeb2* gene has 30 transcripts. According to the structure of *Zeb2* gene, exon4-exon6 of *Zeb2-202* (ENSMUST00000068415.10) transcript is recommended as the knockout region.The region contains 476bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Zeb2* gene. The brief process is as follows:CRISPR/Cas9 system tran 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.

Notice

- According to the existing MGI data,Homozygous null mutants exhibit a variety of defects at embryonic day 8.5 and die between embryonic days 9.5 and 10.5.

- The *Zeb2* gene is located on the Chr2. 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 gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Zeb2 zinc finger E-box binding homeobox 2 [Mus musculus (house mouse)]

Gene ID: 24136, updated on 2-Apr-2019

Summary

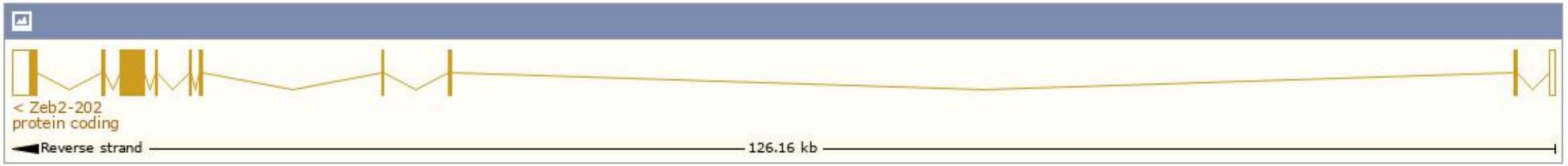
Official Symbol	Zeb2 provided by MGI
Official Full Name	zinc finger E-box binding homeobox 2 provided by MGI
Primary source	MGI:MGI:1344407
See related	Ensembl:ENSMUSG00000026872
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	9130203F04Rik, D130016B08Rik, SIP1, Zfhx1b, Zfx1b, Zfxh1b
Expression	Ubiquitous expression in frontal lobe adult (RPKM 11.4), CNS E18 (RPKM 9.1) and 23 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

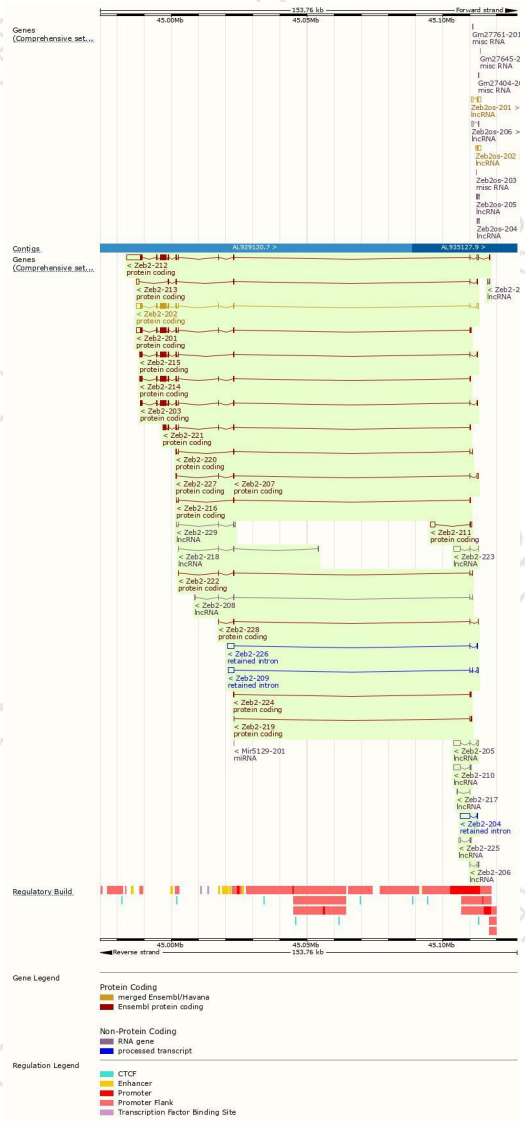
The gene has 30 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Zeb2-212	ENSMUST00000176438.8	9124	1215aa	Protein coding	CCDS16020	Q9R0G7	TSL:5 GENCODE basic APPRIS P2
Zeb2-202	ENSMUST00000068415.10	5624	1215aa	Protein coding	CCDS16020	Q9R0G7	TSL:1 GENCODE basic APPRIS P2
Zeb2-214	ENSMUST00000177302.7	4020	1215aa	Protein coding	CCDS16020	Q9R0G7	TSL:1 GENCODE basic APPRIS P2
Zeb2-201	ENSMUST00000028229.12	5335	1259aa	Protein coding	-	A0A0M3HEP2	TSL:1 GENCODE basic
Zeb2-215	ENSMUST00000200844.3	4064	1191aa	Protein coding	-	A0A0J9YV01	TSL:5 GENCODE basic
Zeb2-203	ENSMUST00000076836.12	3913	1214aa	Protein coding	-	H9H9S3	TSL:5 GENCODE basic APPRIS ALT1
Zeb2-221	ENSMUST00000201804.3	2236	731aa	Protein coding	-	A0A0J9YVF6	CDS 3' incomplete TSL:5
Zeb2-211	ENSMUST00000153561.5	2070	37aa	Protein coding	-	H3BKR1	TSL:1 GENCODE basic
Zeb2-213	ENSMUST00000176732.7	1762	220aa	Protein coding	-	H3BKH5	TSL:5 GENCODE basic
Zeb2-220	ENSMUST00000201623.3	868	214aa	Protein coding	-	A0A0J9YU94	CDS 3' incomplete TSL:5
Zeb2-216	ENSMUST00000201211.3	709	184aa	Protein coding	-	A0A0J9YUY9	CDS 3' incomplete TSL:5
Zeb2-222	ENSMUST00000201969.3	631	141aa	Protein coding	-	A0A0J9YUE5	CDS 3' incomplete TSL:5
Zeb2-207	ENSMUST00000127520.7	577	63aa	Protein coding	-	F6X889	CDS 3' incomplete TSL:5
Zeb2-224	ENSMUST00000202187.3	550	91aa	Protein coding	-	A0A0J9YV93	CDS 3' incomplete TSL:5
Zeb2-219	ENSMUST00000201490.1	462	83aa	Protein coding	-	A0A0J9YU28	CDS 3' incomplete TSL:3
Zeb2-228	ENSMUST00000202935.3	443	113aa	Protein coding	-	A0A0J9YTT5	CDS 3' incomplete TSL:5
Zeb2-227	ENSMUST00000202432.3	240	80aa	Protein coding	-	A0A0J9YUI4	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Zeb2-204	ENSMUST00000123037.2	3852	No protein	Retained intron	-	-	TSL:1
Zeb2-209	ENSMUST00000145529.4	2924	No protein	Retained intron	-	-	TSL:1
Zeb2-226	ENSMUST00000202371.3	2692	No protein	Retained intron	-	-	TSL:1
Zeb2-205	ENSMUST00000124942.4	2989	No protein	lncRNA	-	-	TSL:5
Zeb2-210	ENSMUST00000152232.7	2690	No protein	lncRNA	-	-	TSL:5
Zeb2-223	ENSMUST00000201982.3	2608	No protein	lncRNA	-	-	TSL:5
Zeb2-225	ENSMUST00000202345.1	644	No protein	lncRNA	-	-	TSL:5
Zeb2-208	ENSMUST00000131635.7	618	No protein	lncRNA	-	-	TSL:3
Zeb2-229	ENSMUST00000209076.1	584	No protein	lncRNA	-	-	TSL:5
Zeb2-230	ENSMUST00000238346.1	457	No protein	lncRNA	-	-	
Zeb2-218	ENSMUST00000201413.3	453	No protein	lncRNA	-	-	TSL:5
Zeb2-206	ENSMUST00000126743.1	432	No protein	lncRNA	-	-	TSL:1
Zeb2-217	ENSMUST00000201298.1	370	No protein	lncRNA	-	-	TSL:5

The strategy is based on the design of *Zeb2-202* transcript,The transcription is shown below

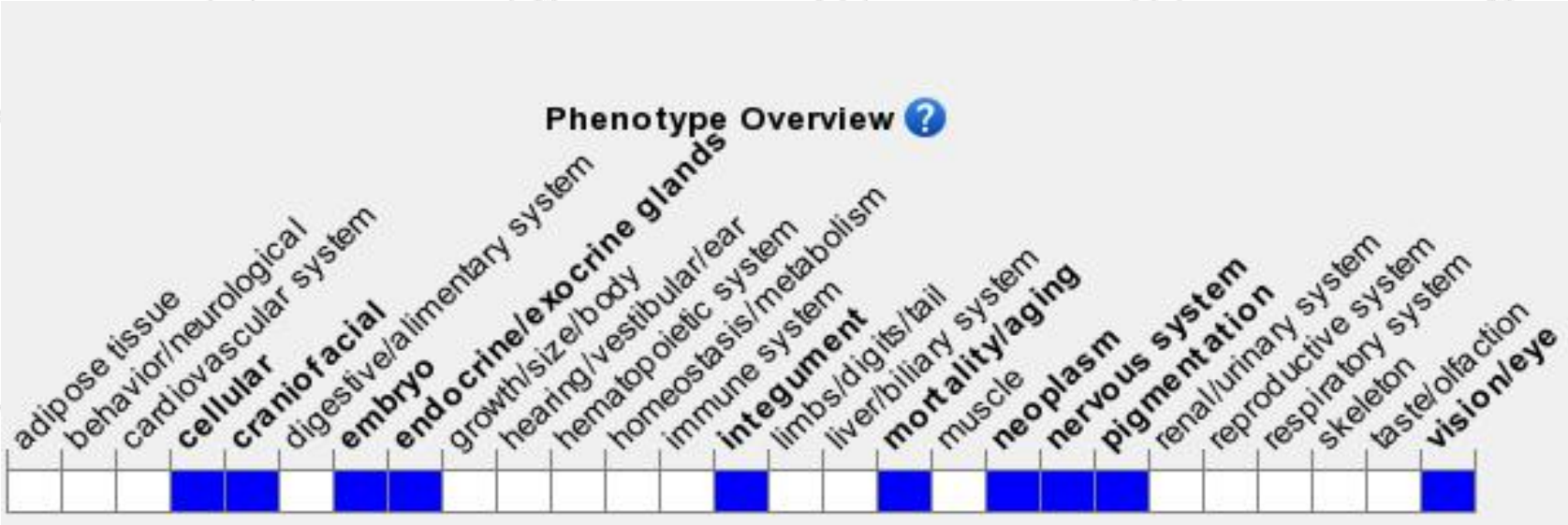


Genomic location distribution



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/).

According to the existing MGI data,Homozygous null mutants exhibit a variety of defects at embryonic day 8.5 and die between embryonic days 9.5 and 10.5.

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

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