Eno2 Cas9-KO Strategy

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Design Date: 2021-5-20

Project Overview

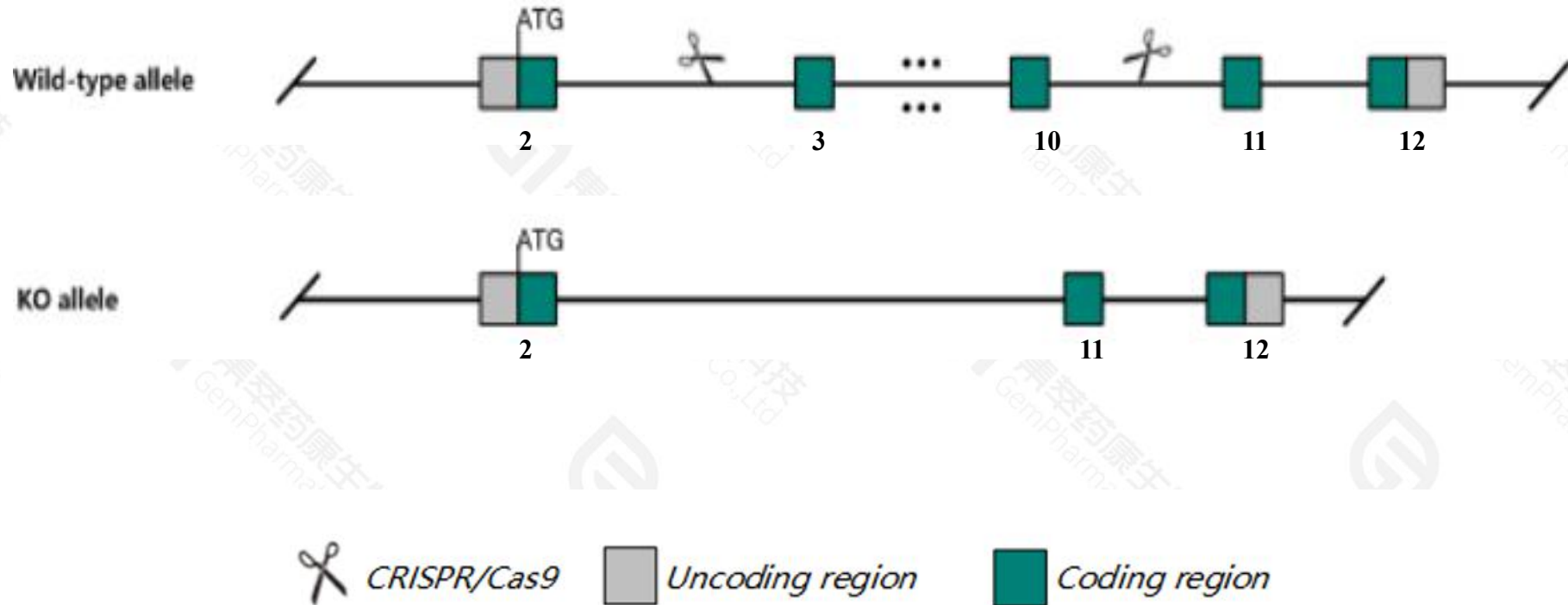|	
Project Name	*Eno2*
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Eno2* gene. The schematic diagram is as follows:

Technical routes

➤ The *Eno2* gene has 16 transcripts. According to the structure of *Eno2* gene, exon3-exon10 of *Eno2-201*(ENSMUST00000004378.15) transcript is recommended as the knockout region. The region contains 1091bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Eno2* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice

➢ The *Eno2* gene is located on the Chr6. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI）

Eno2 enolase 2, gamma neuronal [Mus musculus (house mouse)]

Gene ID: 13807, updated on 13-Mar-2020

▲ Summary

|	
Official Symbol	Eno2 provided by MGI
Official Full Name	enolase 2, gamma neuronal provided by MGI
Primary source	MGI:MGI:95394
See related	Ensembl:ENSMUSG00000004267
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	AI837106, D6Ertd375e, Eno-2, NSE
Expression	Biased expression in cerebellum adult (RPKM 147.0), frontal lobe adult (RPKM 120.3) and 4 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 16 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Eno2-201	ENSMUST00000004378.14	2795	434aa	Protein coding	CCDS20527	P17183 Q545V3	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Eno2-202	ENSMUST00000112476.7	2386	315aa	Protein coding	CCDS80609	D3Z6E4	TSL:1 GENCODE basic
Eno2-216	ENSMUST00000204896.2	1640	391aa	Protein coding	-	A0A0N4SUX5	TSL:5 GENCODE basic
Eno2-213	ENSMUST00000149652.1	804	162aa	Protein coding	-	D3Z2S4	CDS 3' incomplete TSL:2
Eno2-215	ENSMUST00000156033.1	622	58aa	Protein coding	-	A0A0N4SUI6	CDS 3' incomplete TSL:5
Eno2-207	ENSMUST00000135626.7	419	121aa	Protein coding	-	D3YVD3	CDS 3' incomplete TSL:3
Eno2-214	ENSMUST00000151214.7	745	37aa	Nonsense mediated decay	-	A0A0N4SUW8	TSL:5
Eno2-208	ENSMUST00000138770.7	616	37aa	Nonsense mediated decay	-	A0A0N4SUW8	TSL:3
Eno2-204	ENSMUST00000127274.7	565	37aa	Nonsense mediated decay	-	A0A0N4SUW8	TSL:3
Eno2-205	ENSMUST00000128598.1	716	No protein	Processed transcript	-	-	TSL:3
Eno2-212	ENSMUST00000147307.1	2614	No protein	Retained intron	-	-	TSL:2
Eno2-203	ENSMUST00000125436.7	2491	No protein	Retained intron	-	-	TSL:1
Eno2-209	ENSMUST00000139763.7	898	No protein	Retained intron	-	-	TSL:3
Eno2-206	ENSMUST00000128701.7	728	No protein	Retained intron	-	-	TSL:3
Eno2-210	ENSMUST00000146712.1	632	No protein	Retained intron	-	-	TSL:2
Eno2-211	ENSMUST00000147004.1	490	No protein	Retained intron	-	-	TSL:2

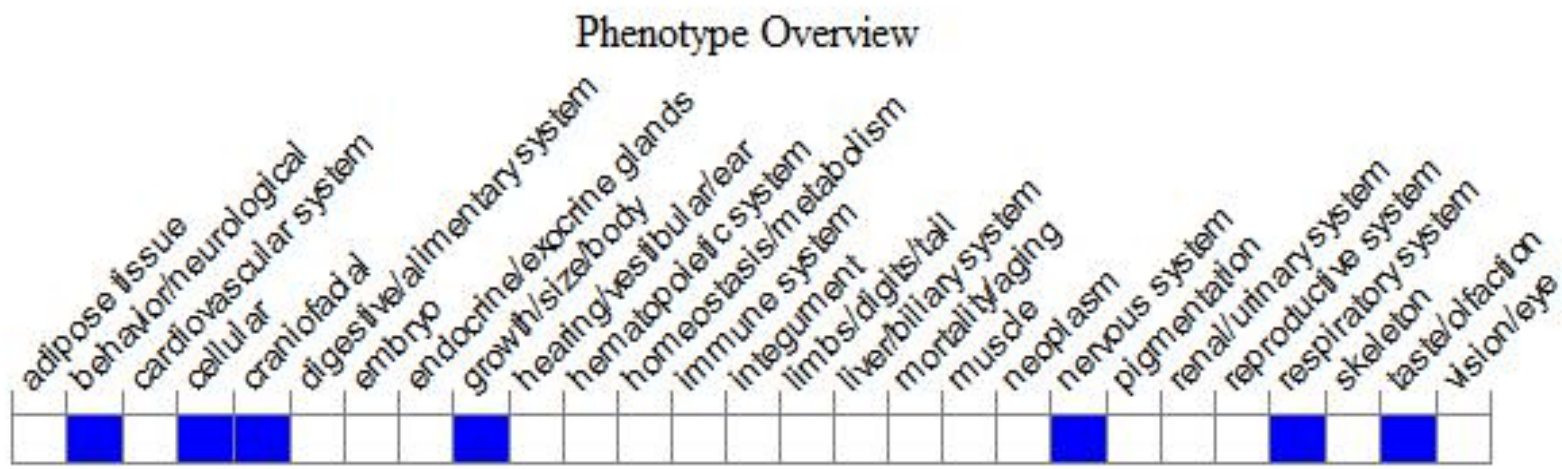The strategy is based on the design of *Eno2-201* transcript,the transcription is shown below:

< Eno2-201
protein coding
◄ Reverse strand

9.62 kb
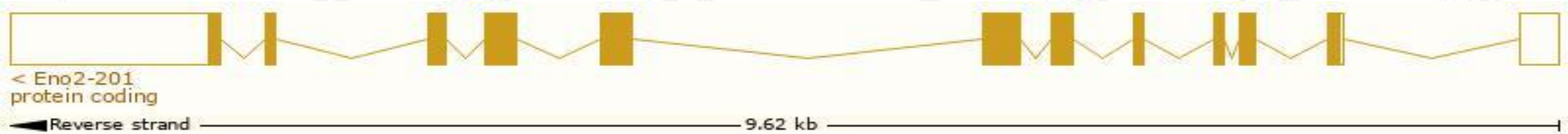
Genomic location distribution
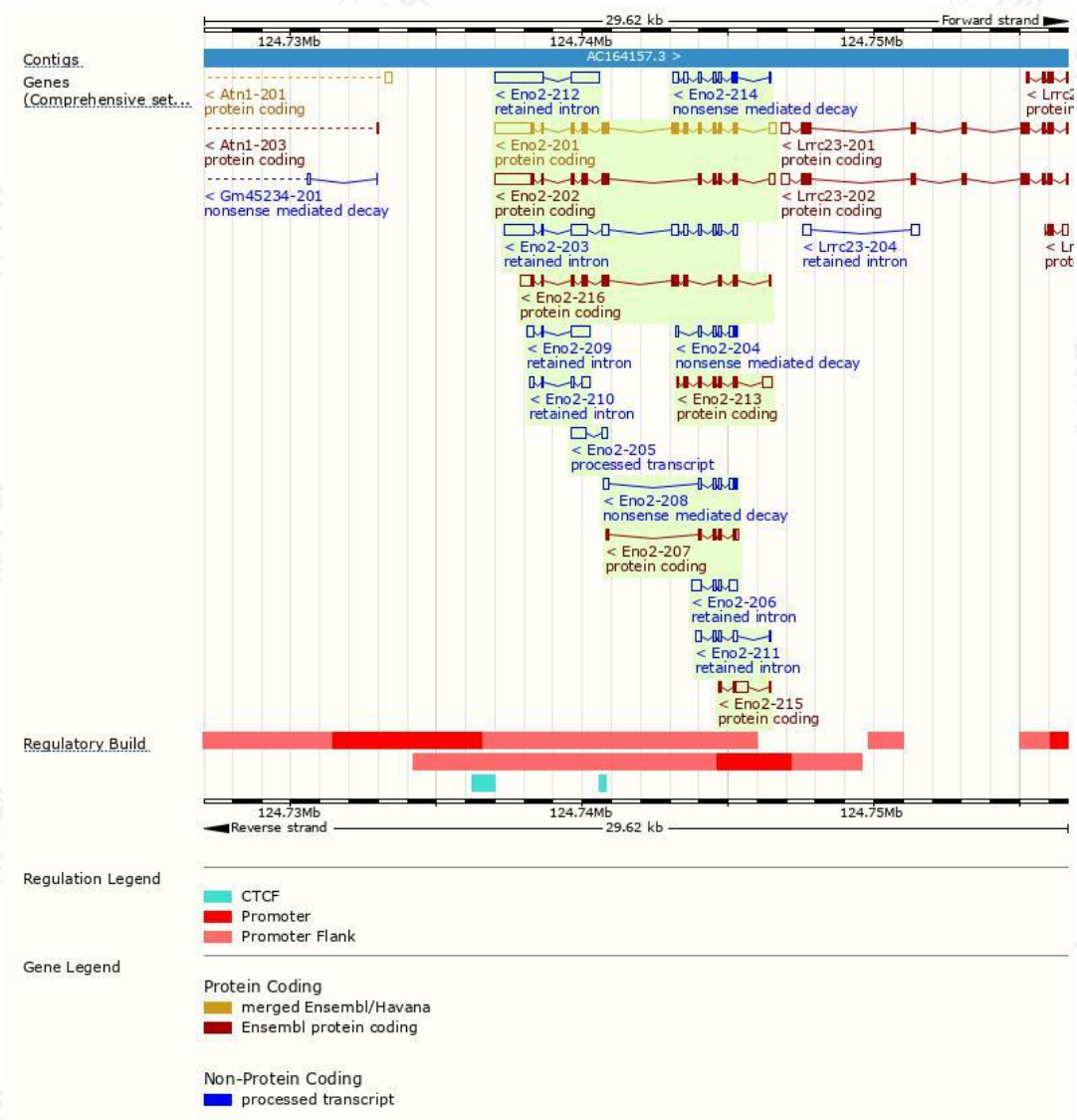
Protein domain
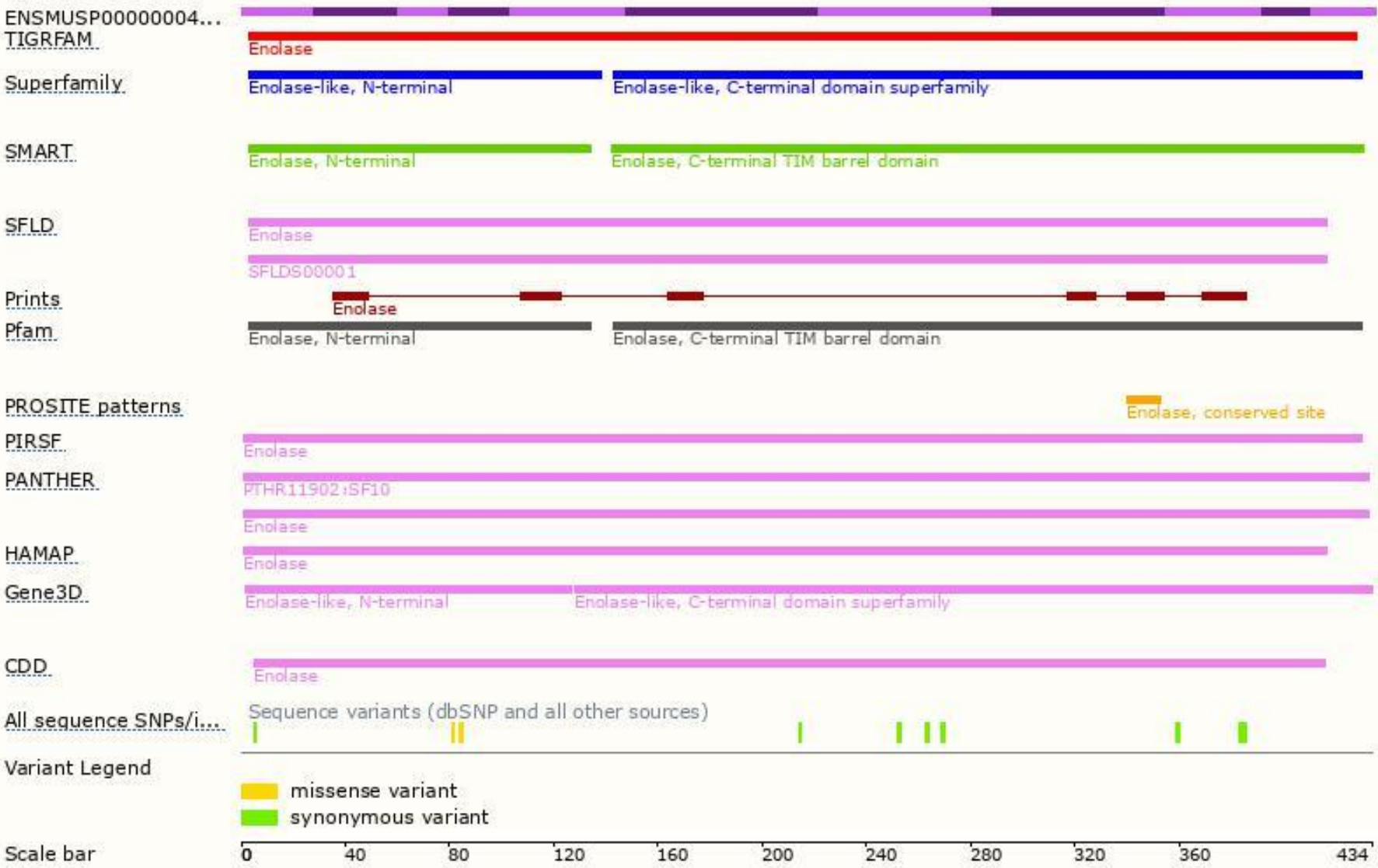
Mouse phenotype description(MGI)

Phenotype Overview

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.
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