

Ap4e1 Cas9-CKO Strategy

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Reviewer:

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Design Date:

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Project Overview

Project Name

Ap4e1

Project type

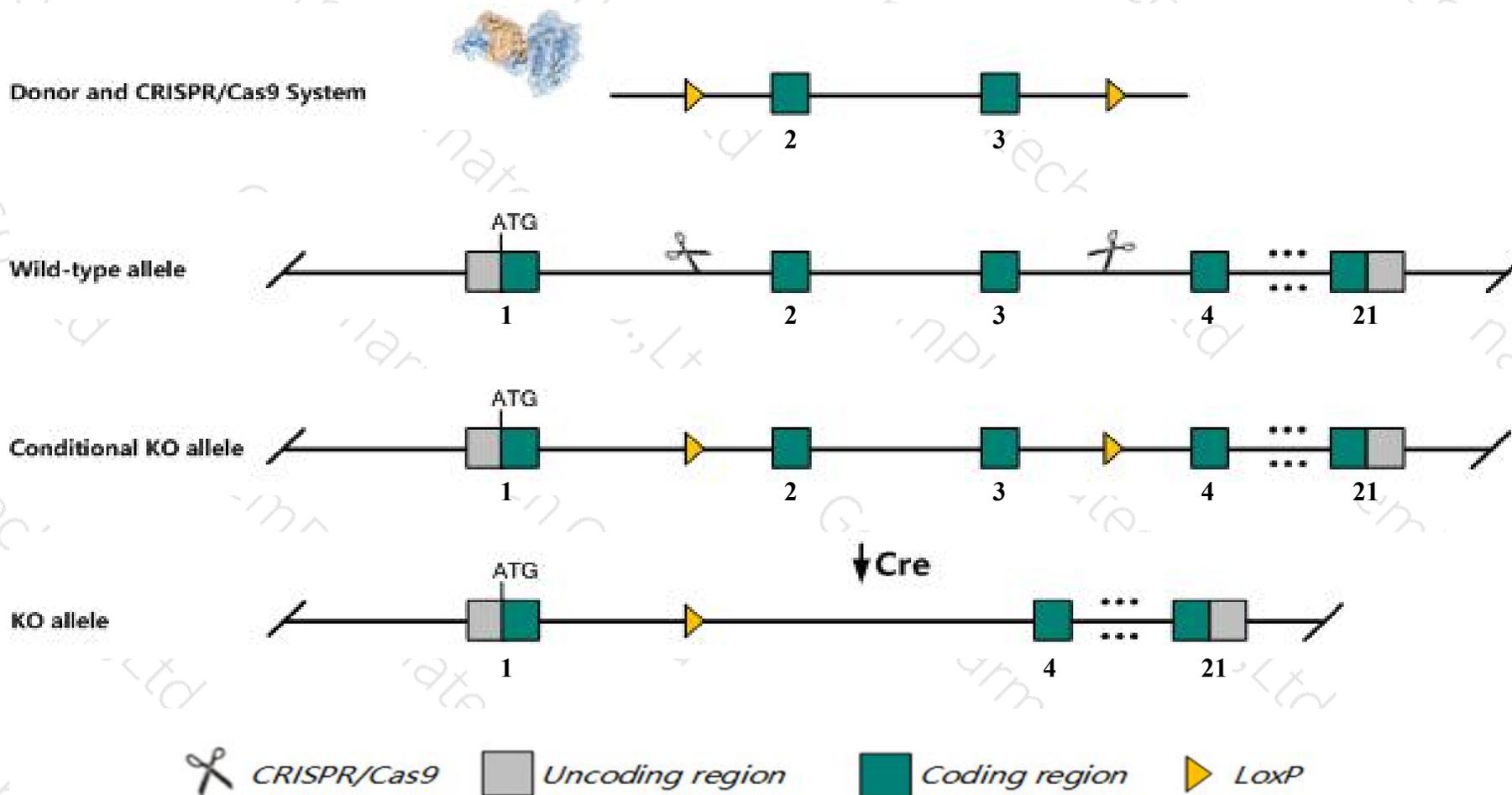
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ap4e1* gene. The schematic diagram is as follows:



- The *Ap4e1* gene has 8 transcripts. According to the structure of *Ap4e1* gene, exon2-exon3 of *Ap4e1-201* (ENSMUST00000002063.14) transcript is recommended as the knockout region. The region contains 196bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ap4e1* 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.

- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit enlarged lateral ventricles, decreased corpus callosum size, decreased vertical activity, and female anemia.
- The *Ap4e1* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Ap4e1 adaptor-related protein complex AP-4, epsilon 1 [Mus musculus (house mouse)]

Gene ID: 108011, updated on 31-Jan-2019

Summary



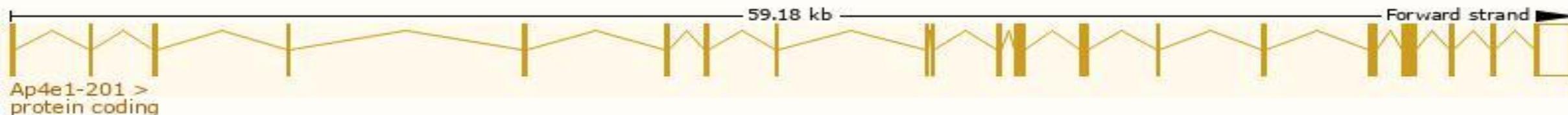
Official Symbol	Ap4e1 provided by MGI
Official Full Name	adaptor-related protein complex AP-4, epsilon 1 provided by MGI
Primary source	MGI:MGI:1336993
See related	Ensembl:ENSMUSG00000001998
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2310033A20Rik, 9930028M04Rik, AV087807
Expression	Ubiquitous expression in CNS E14 (RPKM 3.1), CNS E18 (RPKM 3.0) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

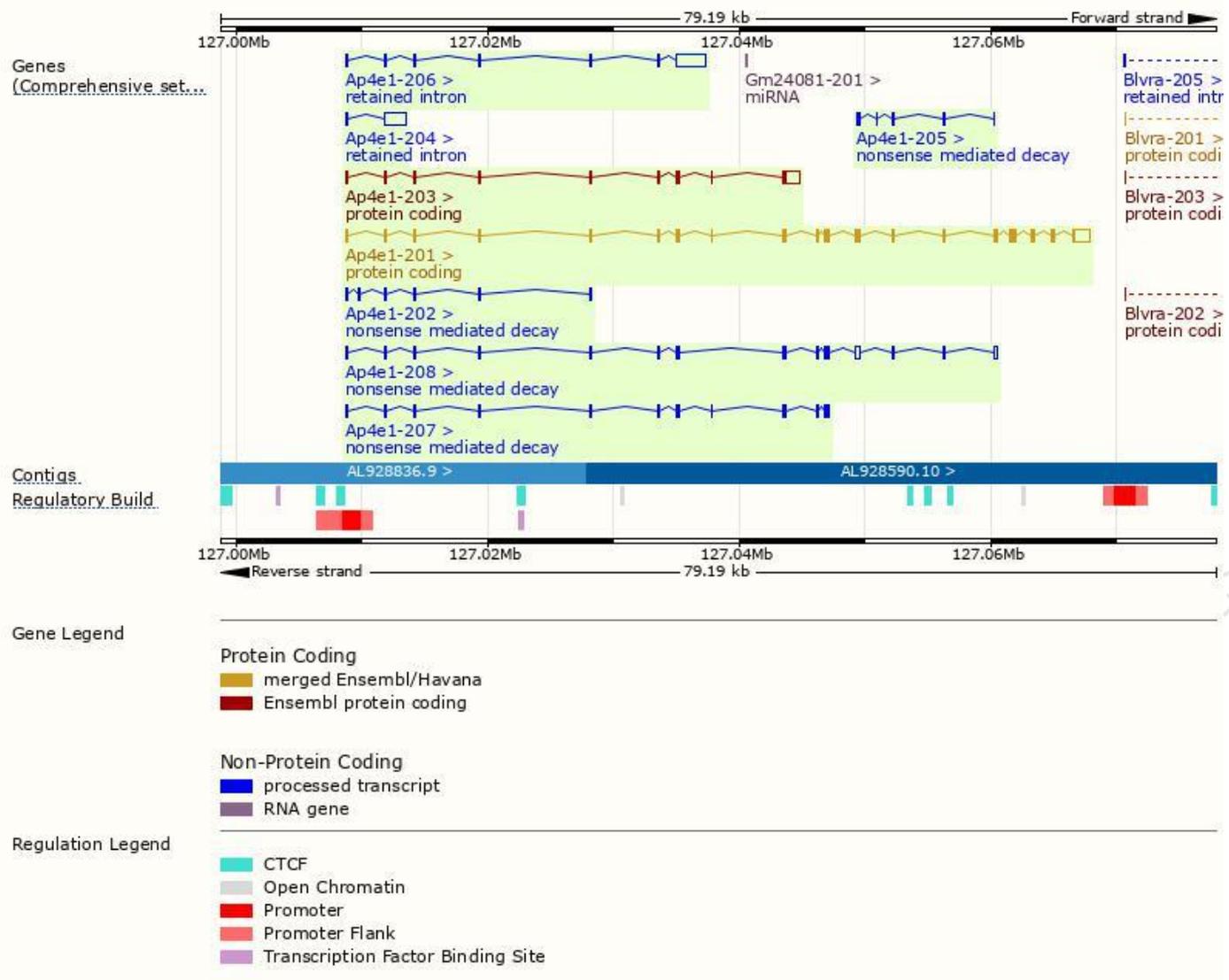
The gene has 8 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ap4e1-201	ENSMUST0000002063.14	4600	1122aa	Protein coding	CCDS38231	Q80V94	TSL:1 GENCODE basic APPRIS P1
Ap4e1-203	ENSMUST00000110394.7	2271	393aa	Protein coding	-	A2ASB3	TSL:1 GENCODE basic
Ap4e1-208	ENSMUST00000177372.7	2272	292aa	Nonsense mediated decay	-	H3BKM4	TSL:5
Ap4e1-207	ENSMUST00000175663.7	1493	367aa	Nonsense mediated decay	-	H3BL03	TSL:5
Ap4e1-202	ENSMUST00000110393.1	731	50aa	Nonsense mediated decay	-	F6W7A9	TSL:2
Ap4e1-205	ENSMUST00000142740.1	659	107aa	Nonsense mediated decay	-	H3BK54	CDS 5' incomplete TSL:5
Ap4e1-206	ENSMUST00000149254.8	3033	No protein	Retained intron	-	-	TSL:5
Ap4e1-204	ENSMUST00000129859.1	1962	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Ap4e1-201* transcript, The transcription is shown below



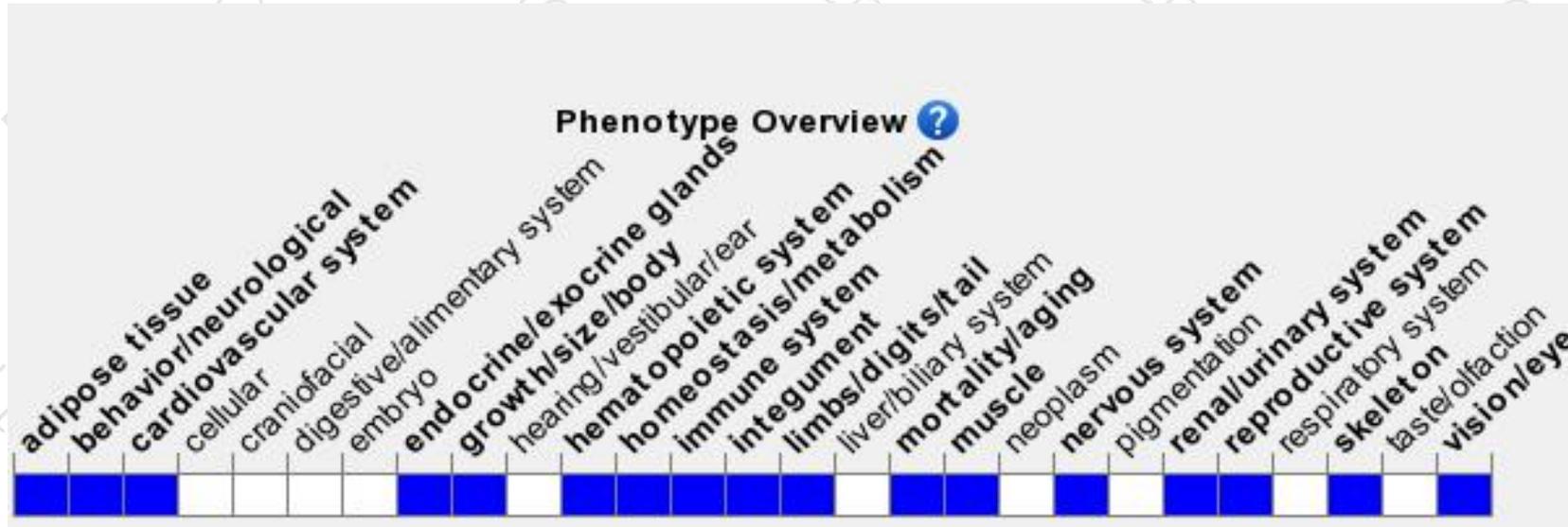
Genomic location distribution



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/>).

According to the existing MGI data, Mice homozygous for a knock-out allele exhibit enlarged lateral ventricles, decreased corpus callosum size, decreased vertical activity, and female anemia.

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

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