

Aopep Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Aopep

Project type

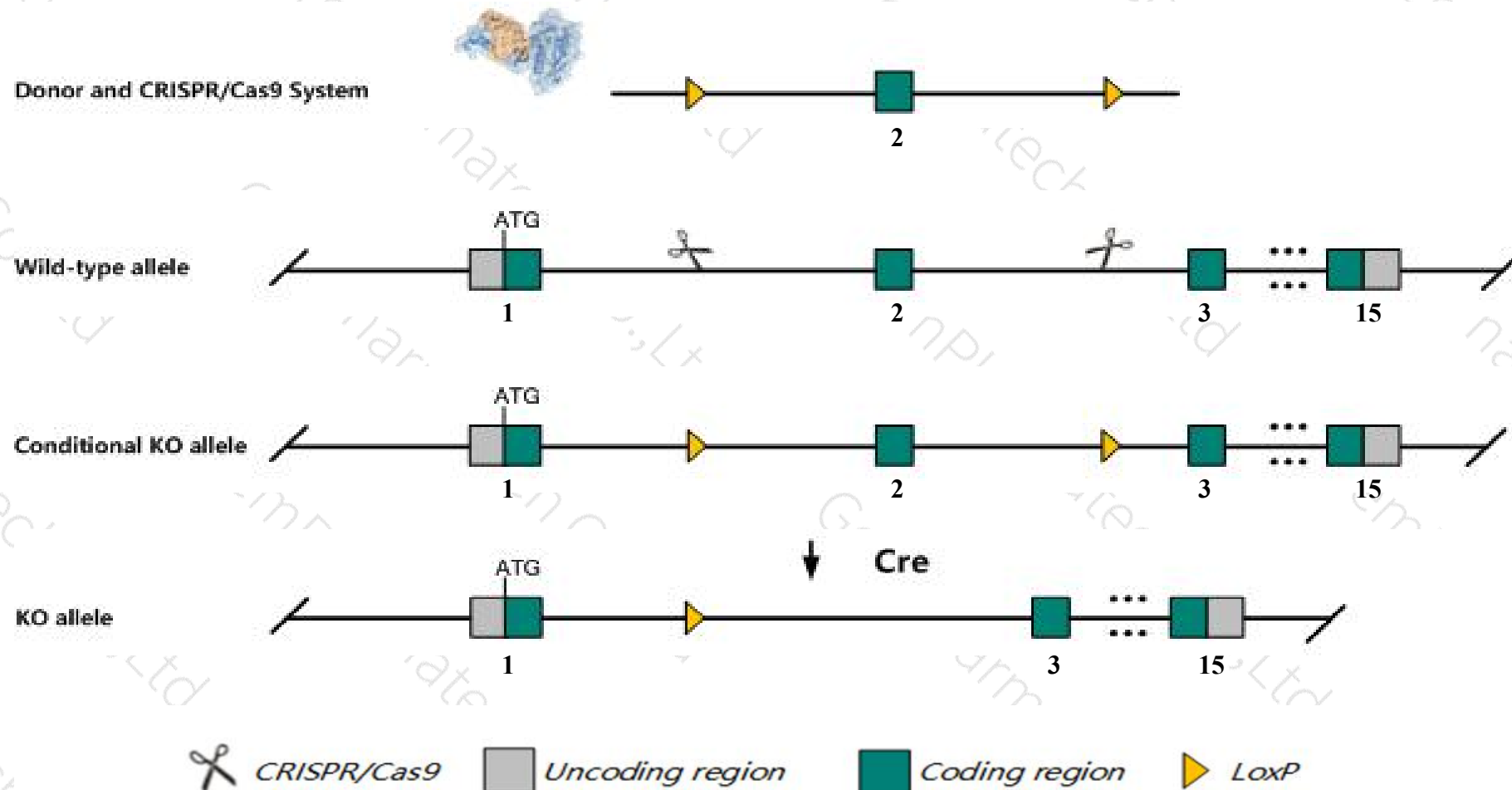
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Aopep* gene has 28 transcripts. According to the structure of *Aopep* gene, exon2 of *Aopep-202* (ENSMUST00000091560.10) transcript is recommended as the knockout region. The region contains 167bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Aopep* 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 one gene trapped allele are phenotypically normal.
- Transcript *Aoep-203, Aoep-216, Aoep-219 and Aoep-224* may not be affected.
- The *Aoep* gene is located on the Chr13. 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)

Aoep aminopeptidase O [Mus musculus (house mouse)]

Gene ID: 72061, updated on 5-Mar-2019

Summary



Official Symbol	Aoep provided by MGI
Official Full Name	aminopeptidase O provided by MGI
Primary source	MGI:MGI:1919311
See related	Ensembl:ENSMUSG00000021458
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	2010111I01Rik, 2300006M17Rik, AP-O, AW742919, ApO, mir-23b, mir-24-1, mir-27b
Expression	Ubiquitous expression in bladder adult (RPKM 14.3), limb E14.5 (RPKM 9.5) and 28 other tissues See more
Orthologs	human all

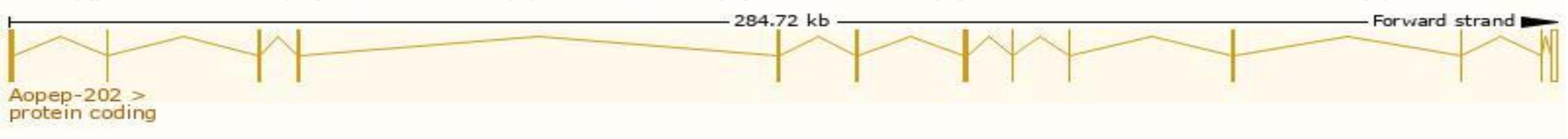
Transcript information（Ensembl）



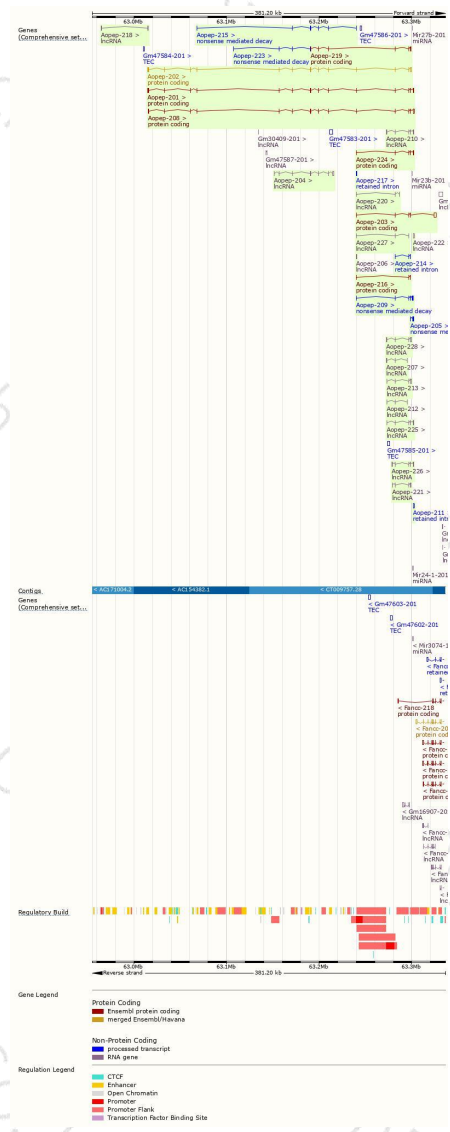
The gene has 28 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Aoep-202	ENSMUST00000091560.10	3509	823aa	Protein coding	CCDS36699	Q3UQZ7	TSL1 GENCODE basic APPRIS P2
Aoep-208	ENSMUST00000220863.1	9249	715aa	Protein coding	-	A0A1Y7VJQ8	CDS 5' incomplete TSL1
Aoep-201	ENSMUST00000021911.14	2979	822aa	Protein coding	-	F8WGB2	TSL5 GENCODE basic APPRIS ALT2
Aoep-203	ENSMUST00000159152.2	2824	167aa	Protein coding	-	F6SS74	CDS 5' incomplete TSL1
Aoep-224	ENSMUST00000222929.1	1055	182aa	Protein coding	-	A0A1Y7VN88	CDS 5' incomplete TSL1
Aoep-219	ENSMUST00000222282.1	776	170aa	Protein coding	-	A0A1Y7VLR8	CDS 5' incomplete TSL2
Aoep-216	ENSMUST00000221820.1	331	110aa	Protein coding	-	A0A1Y7VLP8	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL5
Aoep-209	ENSMUST00000220884.1	772	155aa	Nonsense mediated decay	-	A0A1Y7VLB5	CDS 5' incomplete TSL5
Aoep-215	ENSMUST00000221676.1	718	69aa	Nonsense mediated decay	-	A0A1Y7VJX1	CDS 5' incomplete TSL3
Aoep-223	ENSMUST00000222907.1	539	67aa	Nonsense mediated decay	-	A0A1Y7VLV4	CDS 5' incomplete TSL3
Aoep-205	ENSMUST00000220485.1	493	19aa	Nonsense mediated decay	-	A0A1Y7VNZ9	CDS 5' incomplete TSL3
Aoep-211	ENSMUST00000221108.1	1599	No protein	Retained intron	-	-	TSLNA
Aoep-214	ENSMUST00000221501.1	587	No protein	Retained intron	-	-	TSL2
Aoep-217	ENSMUST00000221938.1	268	No protein	Retained intron	-	-	TSL3
Aoep-204	ENSMUST00000220457.1	1754	No protein	lncRNA	-	-	TSL1
Aoep-228	ENSMUST00000223204.1	1754	No protein	lncRNA	-	-	TSL1
Aoep-213	ENSMUST00000221364.1	1481	No protein	lncRNA	-	-	TSL1
Aoep-226	ENSMUST00000223073.1	1376	No protein	lncRNA	-	-	TSL2
Aoep-221	ENSMUST00000222721.1	1007	No protein	lncRNA	-	-	TSL2
Aoep-222	ENSMUST00000222646.1	921	No protein	lncRNA	-	-	TSL2
Aoep-225	ENSMUST00000223007.1	918	No protein	lncRNA	-	-	TSL5
Aoep-210	ENSMUST00000220885.1	894	No protein	lncRNA	-	-	TSL3
Aoep-207	ENSMUST00000220670.1	688	No protein	lncRNA	-	-	TSL1
Aoep-218	ENSMUST00000222181.1	677	No protein	lncRNA	-	-	TSL2
Aoep-212	ENSMUST00000221164.1	459	No protein	lncRNA	-	-	TSL5
Aoep-206	ENSMUST00000220563.1	424	No protein	lncRNA	-	-	TSL2
Aoep-220	ENSMUST00000222680.1	353	No protein	lncRNA	-	-	TSL3
Aoep-227	ENSMUST00000223185.1	348	No protein	lncRNA	-	-	TSL5

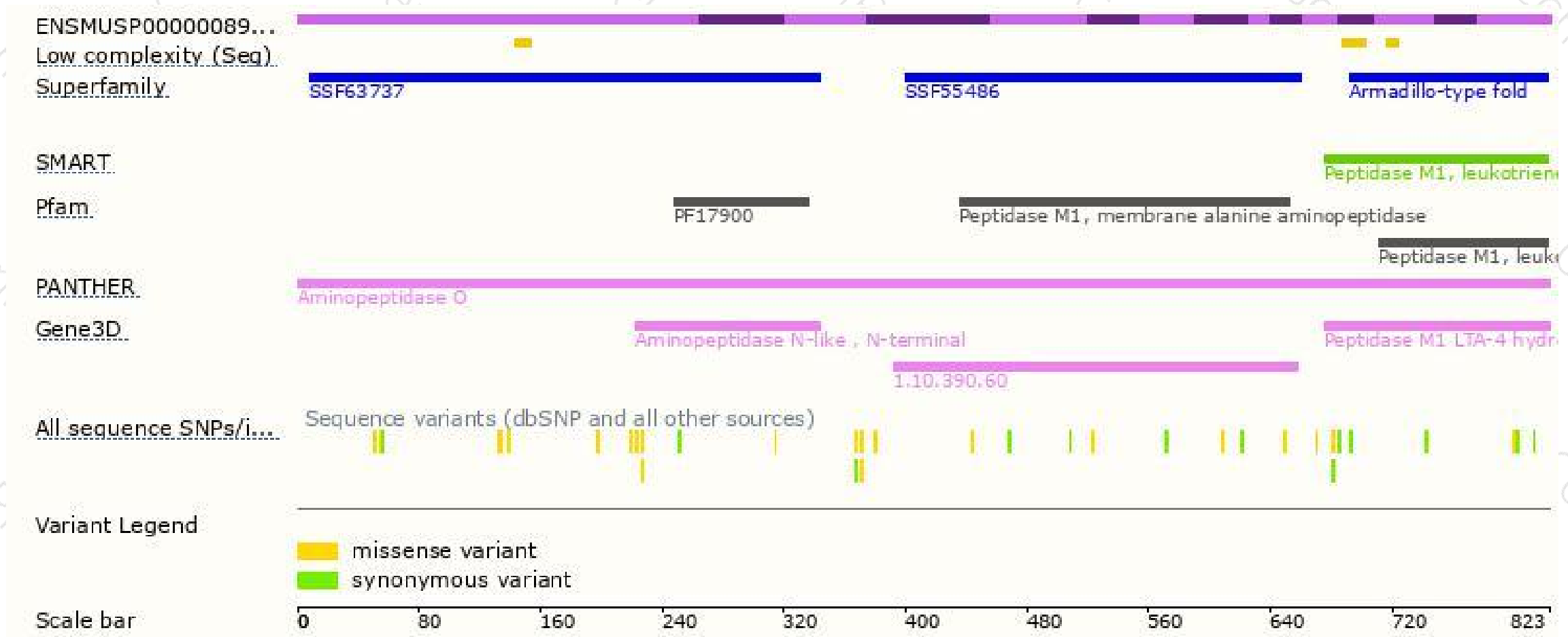
The strategy is based on the design of *Aoep-202* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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