

Alox3 Cas9-CKO Strategy

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Project Overview

Project Name

Aloxe3

Project type

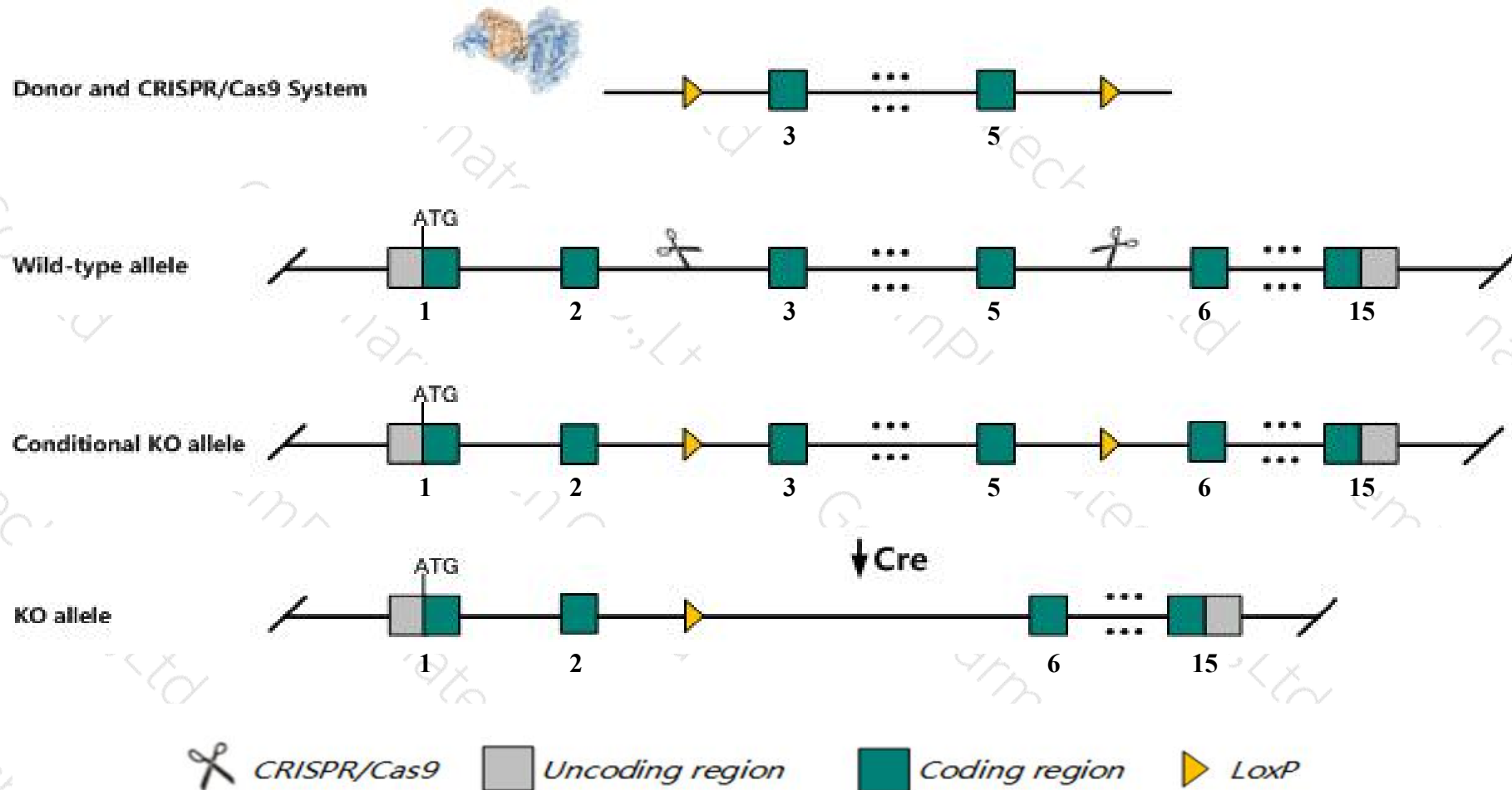
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Alox3* gene. The schematic diagram is as follows:



Technical routes

- The *Aloxe3* gene has 6 transcripts. According to the structure of *Aloxe3* gene, exon3-exon5 of *Aloxe3-201* (ENSMUST00000021268.8) transcript is recommended as the knockout region. The region contains 328bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Aloxe3* 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 complete neonatal lethality, impaired skin barrier function, dehydration, tightly packed stratum corneum, impaired stratum corneum desquamation and reduced levels of ester-bound ceramide in the epidermis.
- The *Aloxe3* gene is located on the Chr11. 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)

Aloxe3 arachidonate lipoxygenase 3 [Mus musculus (house mouse)]

Gene ID: 23801, updated on 19-Feb-2019

Summary



Official Symbol	Aloxe3 provided by MGI
Official Full Name	arachidonate lipoxygenase 3 provided by MGI
Primary source	MGI:MGI:1345140
See related	Ensembl:ENSMUSG00000020892
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	e-LOX-3, eLOX-3
Expression	Biased expression in stomach adult (RPKM 6.6), CNS E18 (RPKM 2.7) and 10 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

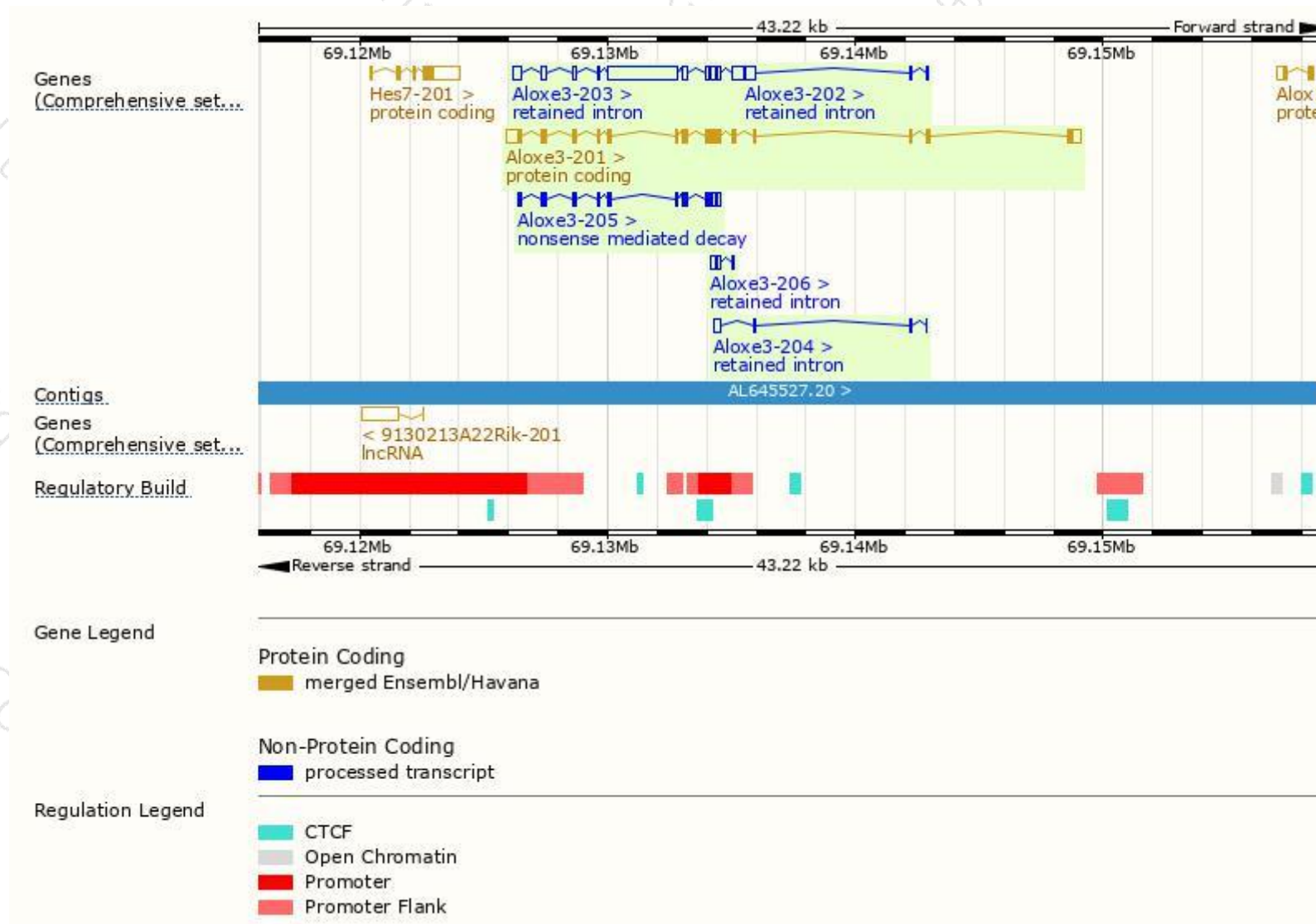
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Aloxe3-201	ENSMUST00000021268.8	3019	711aa	Protein coding	CCDS24884	Q9WV07	TSL:1 GENCODE basic APPRIS P1
Aloxe3-205	ENSMUST00000175661.1	1368	380aa	Nonsense mediated decay	-	H3BJ21	TSL:5
Aloxe3-203	ENSMUST00000155324.8	4684	No protein	Retained intron	-	-	TSL:2
Aloxe3-202	ENSMUST00000139257.1	638	No protein	Retained intron	-	-	TSL:3
Aloxe3-204	ENSMUST00000156874.7	454	No protein	Retained intron	-	-	TSL:5
Aloxe3-206	ENSMUST00000176087.1	414	No protein	Retained intron	-	-	TSL:2

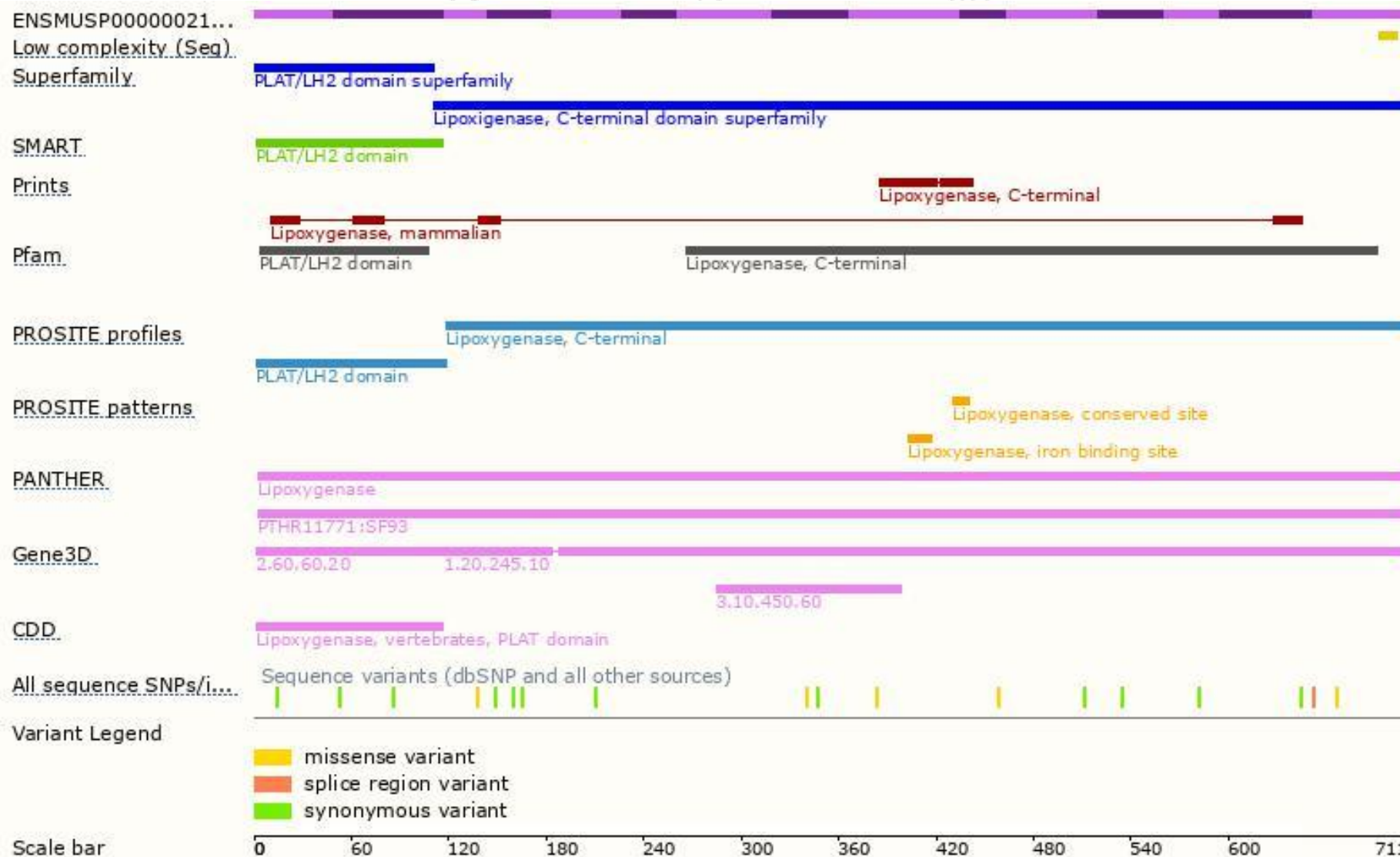
The strategy is based on the design of *Aloxe3-201* transcript,The transcription is shown below



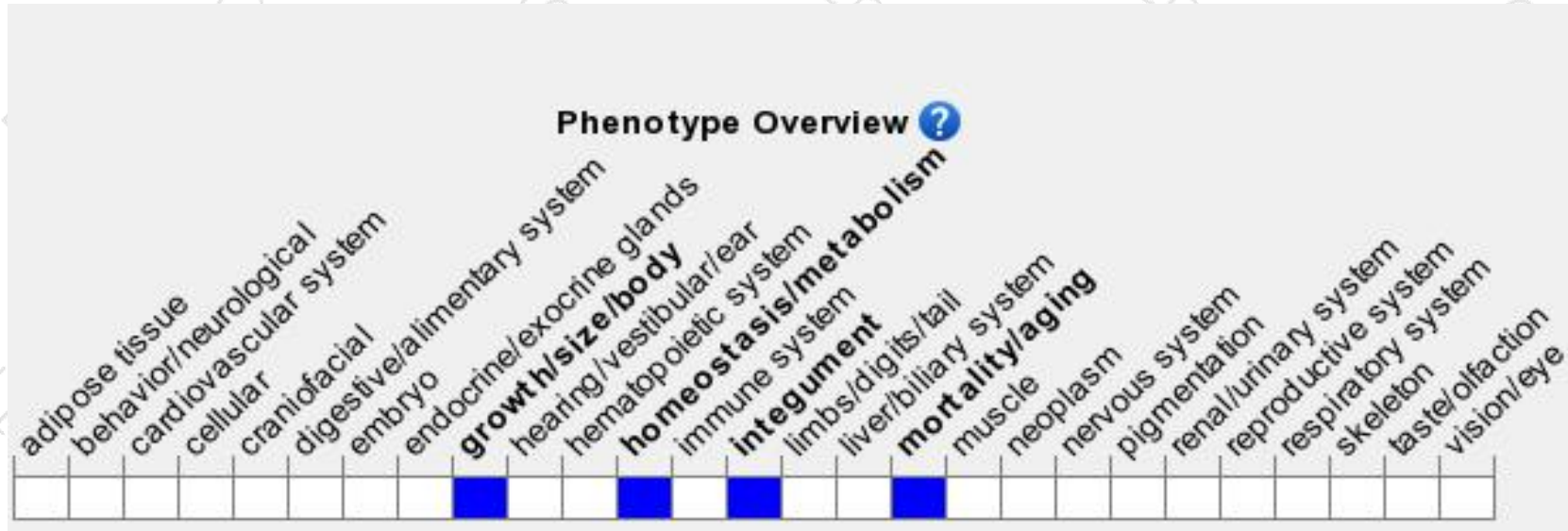
Genomic location distribution



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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