

Idh3a Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Idh3a

Project type

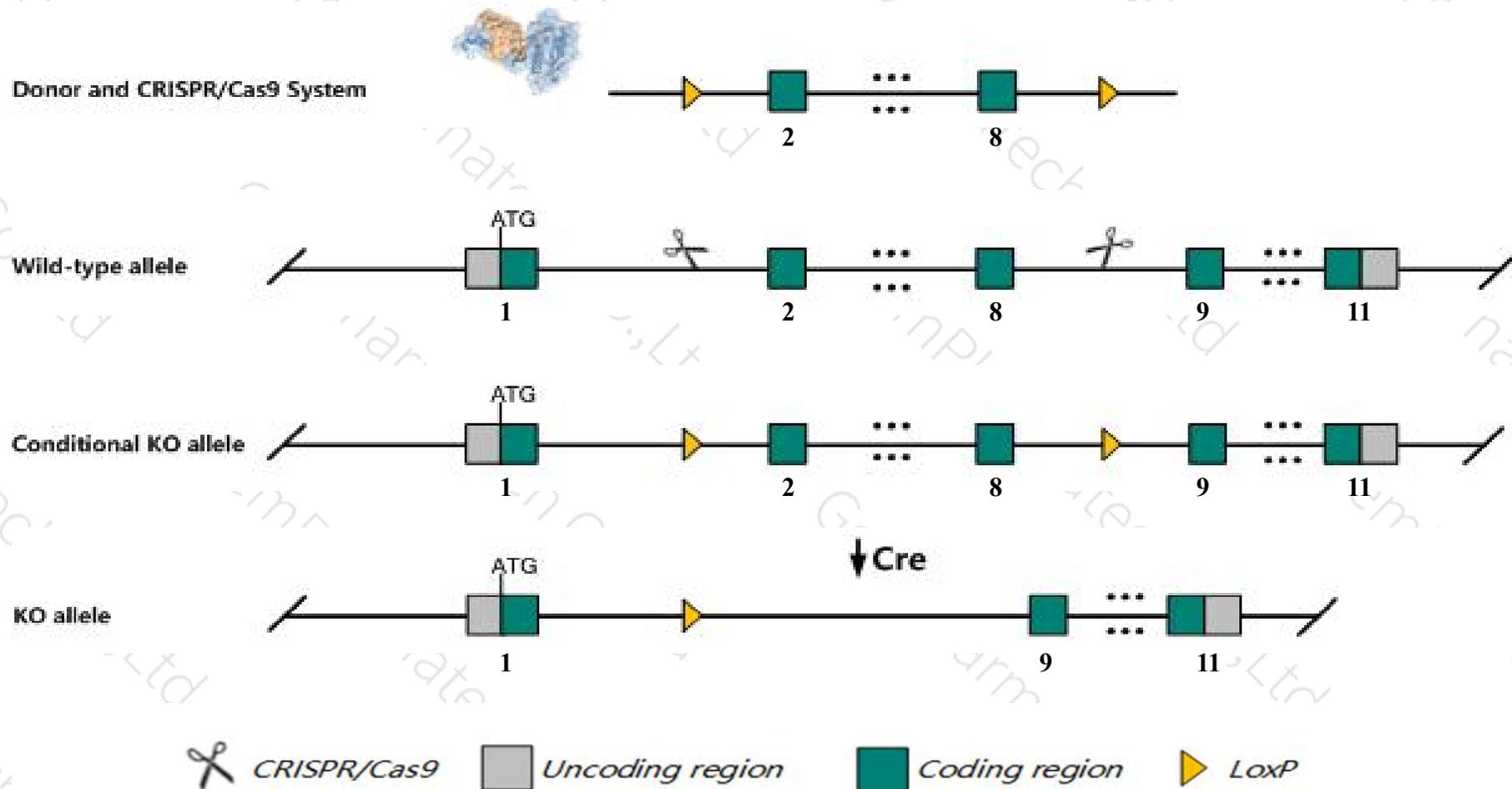
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Idh3a* gene. The schematic diagram is as follows:



- The *Idh3a* gene has 5 transcripts. According to the structure of *Idh3a* gene, exon2-exon8 of *Idh3a-201* (ENSMUST00000167866.1) transcript is recommended as the knockout region. The region contains 752bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Idh3a* 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 an ENU-induced mutation exhibit progressive retinal degeneration and decreased visual acuity.
- The *Idh3a* gene is located on the Chr9. 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)

Idh3a isocitrate dehydrogenase 3 (NAD+) alpha [*Mus musculus* (house mouse)]

Gene ID: 67834, updated on 27-Aug-2019

Summary

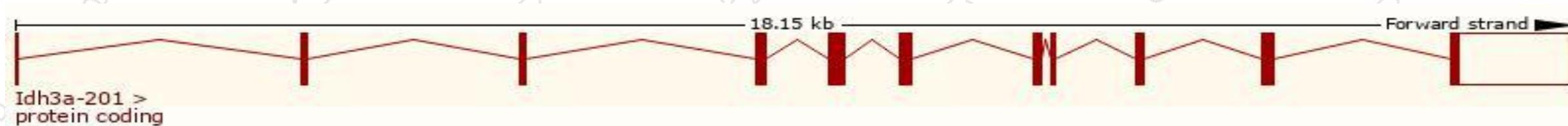
Official Symbol	Idh3a provided by MGI
Official Full Name	isocitrate dehydrogenase 3 (NAD+) alpha provided by MGI
Primary source	MGI:MGI:1915084
See related	Ensembl:ENSMUSG00000032279
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AA407078; AI316514; 1110003P10Rik; 1500012E04Rik
Expression	Ubiquitous expression in heart adult (RPKM 153.2), cerebellum adult (RPKM 51.8) and 25 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

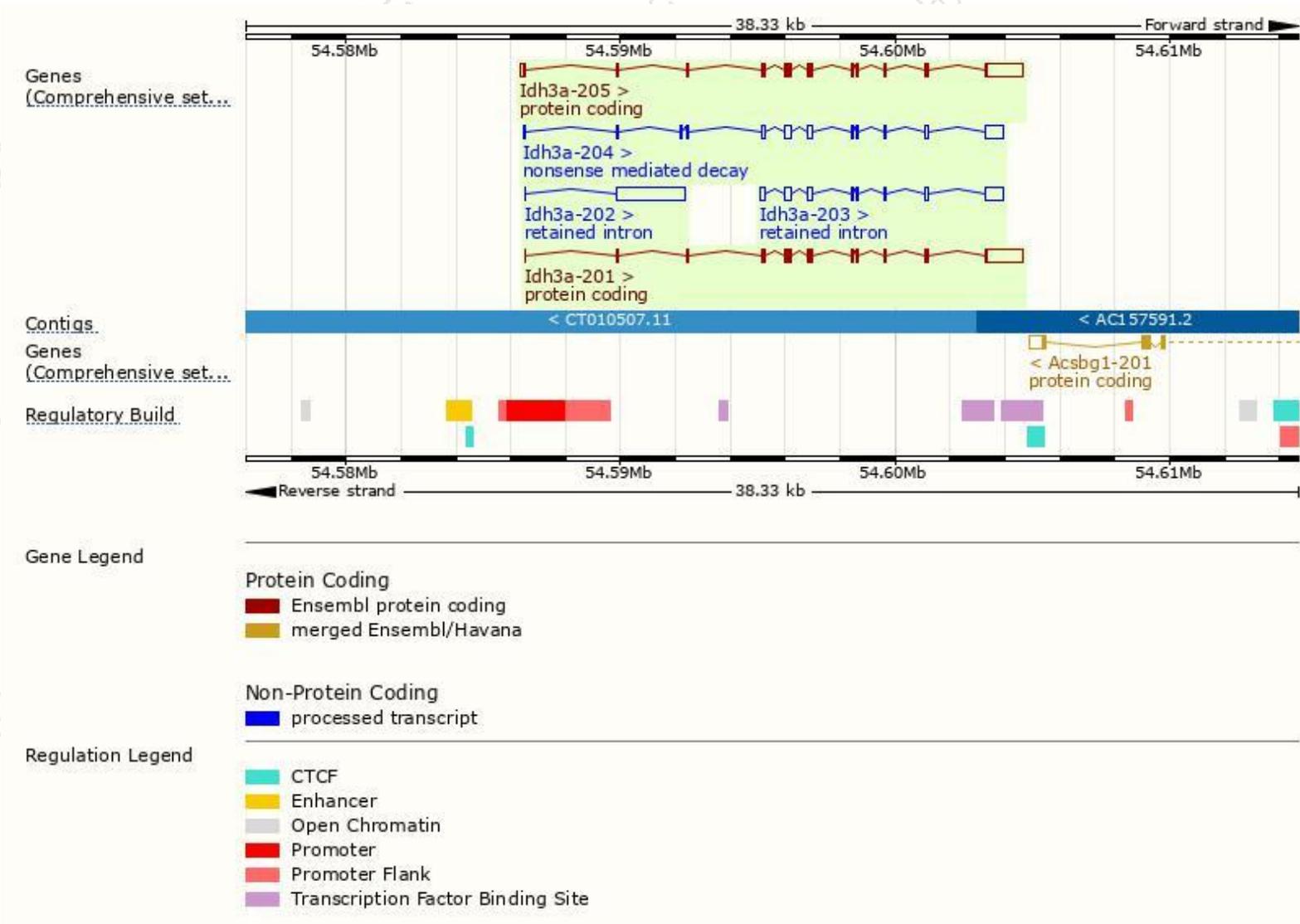
The gene has 5 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Idh3a-201	ENSMUST00000167866.1	2403	366aa	Protein coding	CCDS23191	Q9D6R2	TSL:1 GENCODE basic APPRIS P1
Idh3a-205	ENSMUST00000217484.1	2580	384aa	Protein coding	-	A0A1L1STE6	TSL:1 GENCODE basic
Idh3a-204	ENSMUST00000215273.1	1812	37aa	Nonsense mediated decay	-	A0A1L1SQF9	TSL:1
Idh3a-202	ENSMUST00000213729.1	2517	No protein	Retained intron	-	-	TSL:1
Idh3a-203	ENSMUST00000214136.1	1570	No protein	Retained intron	-	-	TSL:1

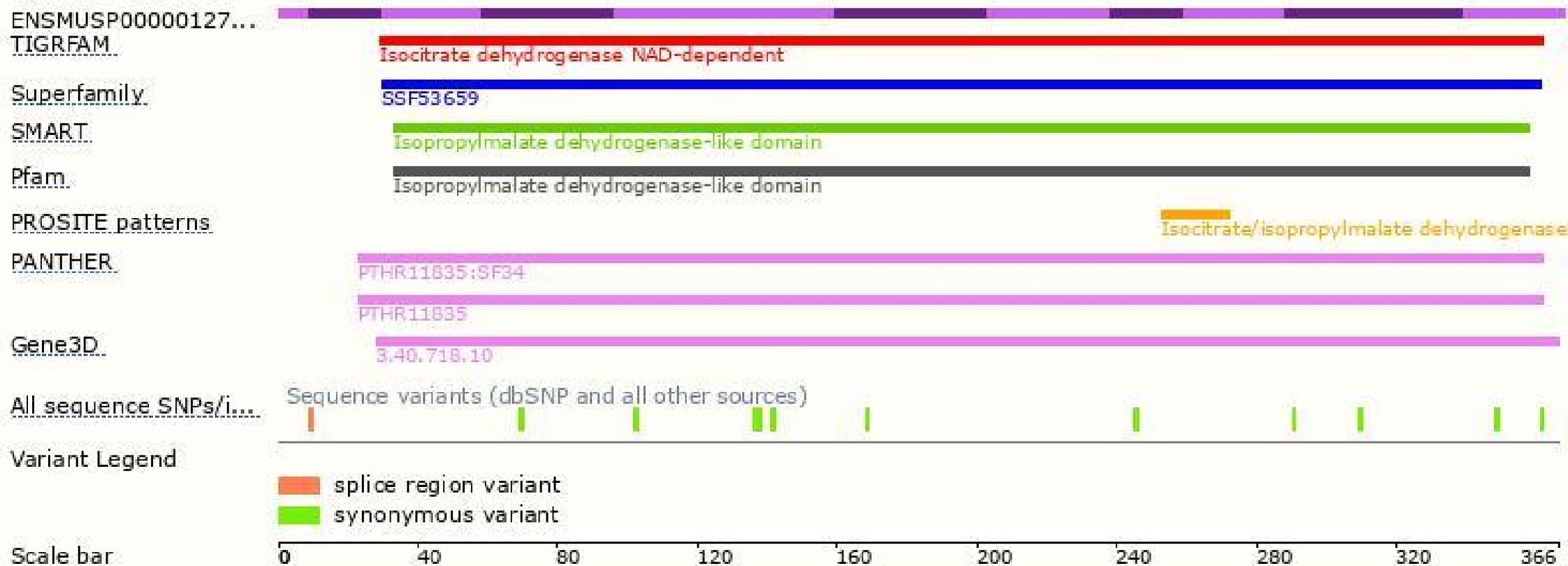
The strategy is based on the design of *Idh3a-201* transcript, The transcription is shown below



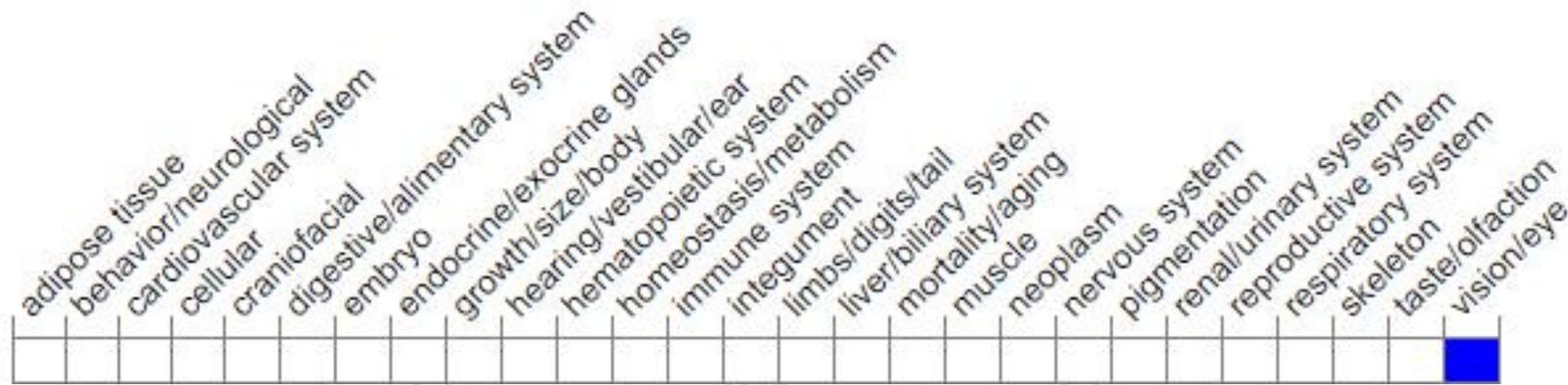
Genomic location distribution



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 an ENU-induced mutation exhibit progressive retinal degeneration and decreased visual acuity.

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

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