

Notch4 Cas9-CKO Strategy

Designer:

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Project Overview

Project Name

Notch4

Project type

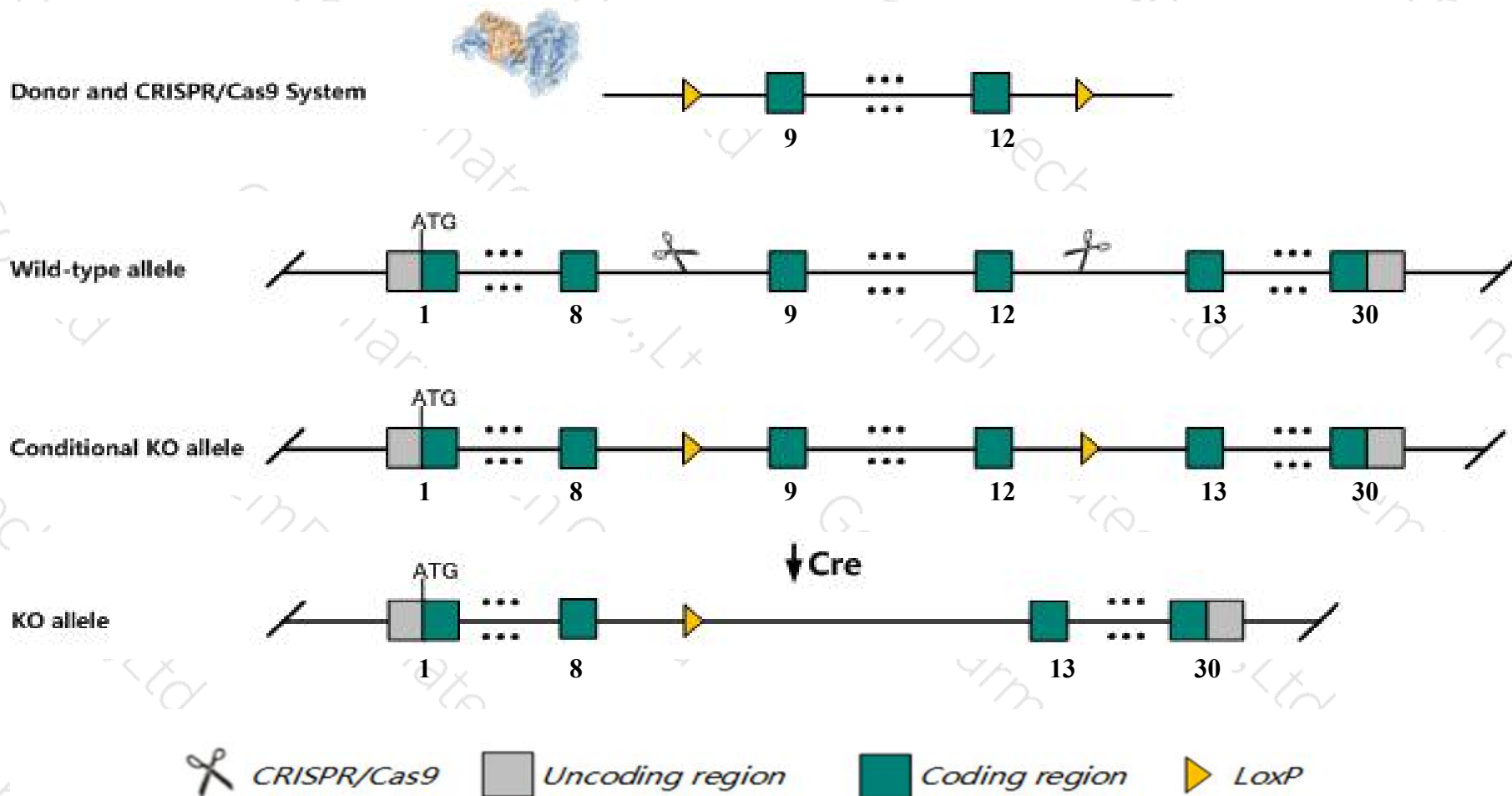
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Notch4* gene. The schematic diagram is as follows:



Technical routes

- The *Notch4* gene has 12 transcripts. According to the structure of *Notch4* gene, exon9-exon12 of *Notch4-201* (ENSMUST00000015612.13) transcript is recommended as the knockout region. The region contains 511bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Notch4* 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 are viable and fertile but exhibit a slight delay in postnatal retinal angiogenesis.
- The *Notch4* gene is located on the Chr17. 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)

Notch4 notch 4 [Mus musculus (house mouse)]

Gene ID: 18132, updated on 31-Jan-2019

Summary



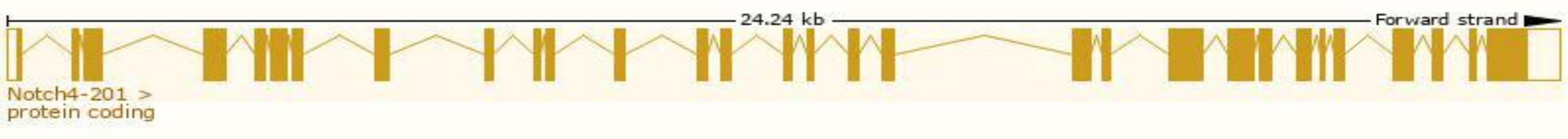
Official Symbol	Notch4 provided by MGI
Official Full Name	notch 4 provided by MGI
Primary source	MGI:MGI:107471
See related	Ensembl:ENSMUSG00000015468
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Int-3, Int3, N4
Expression	Biased expression in lung adult (RPKM 37.2), adrenal adult (RPKM 14.3) and 14 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

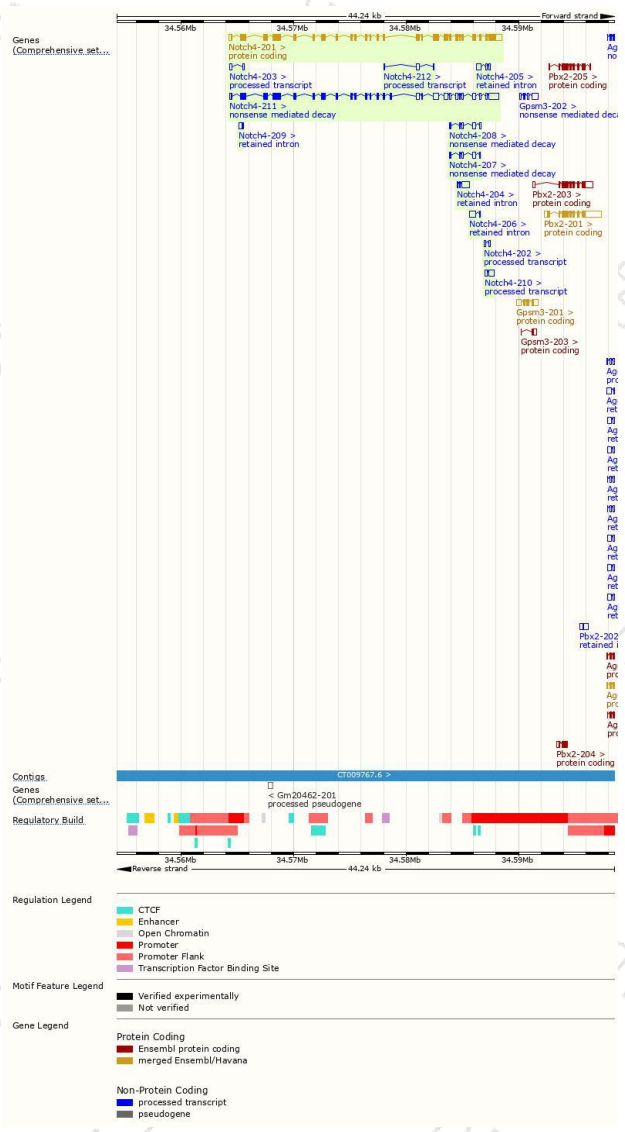
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Notch4-201	ENSMUST00000015612.13	6591	1964aa	Protein coding	CCDS28647	A2CG28	TSL:1 GENCODE basic APPRIS P1
Notch4-211	ENSMUST000000173389.7	6380	978aa	Nonsense mediated decay	-	G3UX69	TSL:1
Notch4-208	ENSMUST000000151867.1	804	43aa	Nonsense mediated decay	-	G3UZH3	CDS 5' incomplete TSL:5
Notch4-207	ENSMUST000000151654.7	733	43aa	Nonsense mediated decay	-	G3UZH3	CDS 5' incomplete TSL:5
Notch4-210	ENSMUST000000156724.1	619	No protein	Processed transcript	-	-	TSL:2
Notch4-212	ENSMUST000000174707.1	451	No protein	Processed transcript	-	-	TSL:3
Notch4-203	ENSMUST000000126950.1	356	No protein	Processed transcript	-	-	TSL:3
Notch4-202	ENSMUST000000126702.1	247	No protein	Processed transcript	-	-	TSL:5
Notch4-204	ENSMUST000000128314.1	796	No protein	Retained intron	-	-	TSL:3
Notch4-206	ENSMUST000000150441.1	642	No protein	Retained intron	-	-	TSL:5
Notch4-205	ENSMUST000000141768.1	598	No protein	Retained intron	-	-	TSL:2
Notch4-209	ENSMUST000000152714.1	352	No protein	Retained intron	-	-	TSL:3

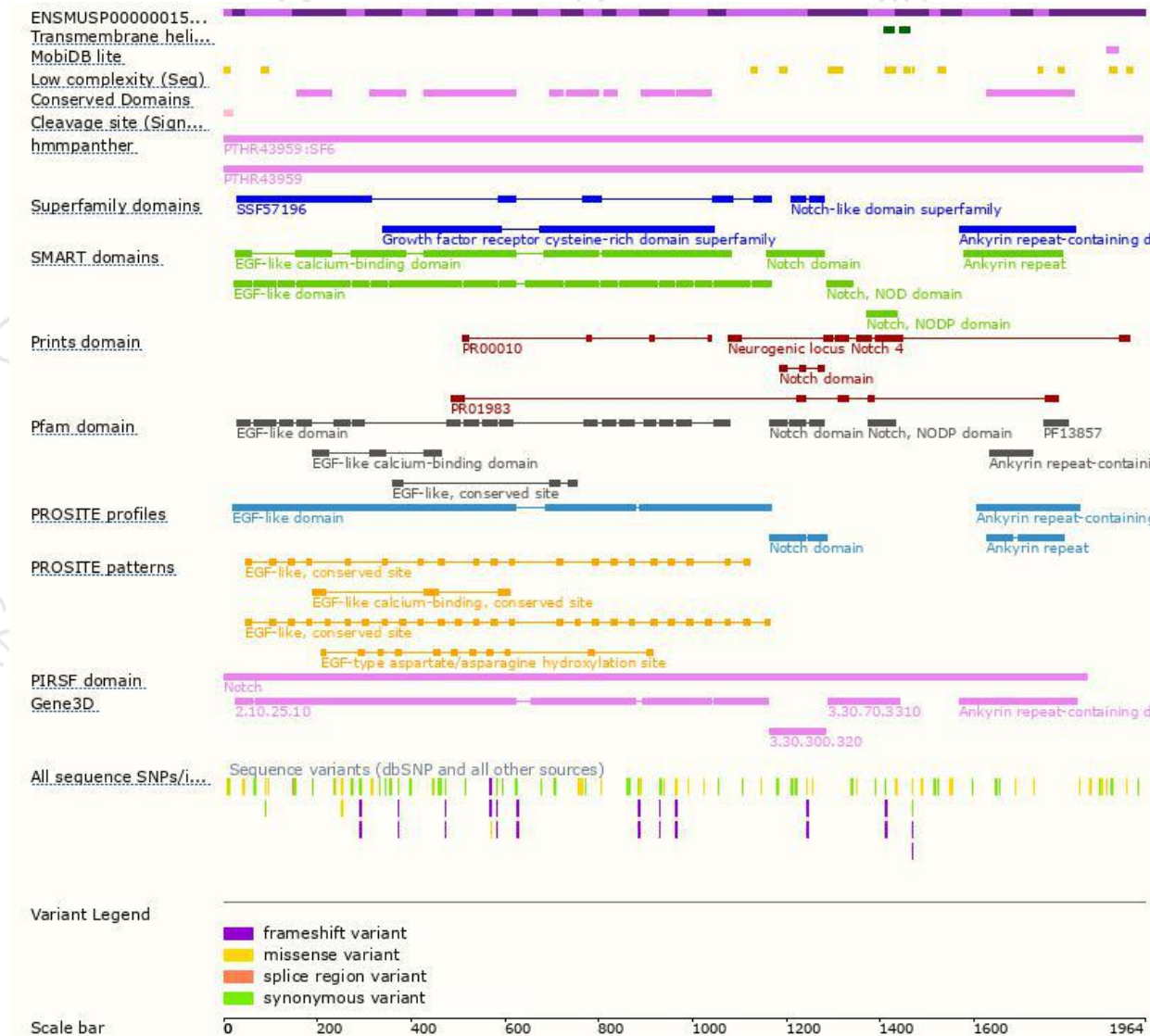
The strategy is based on the design of *Notch4-201* transcript,The transcription is shown below



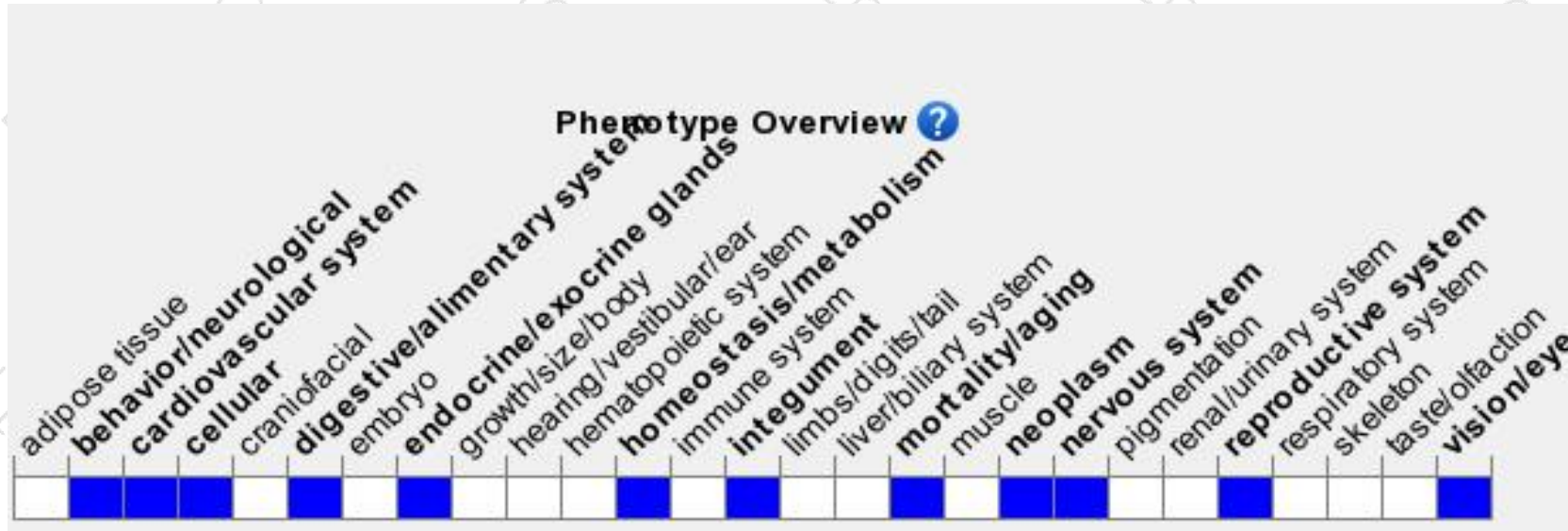
Genomic location distribution



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 are viable and fertile but exhibit a slight delay in postnatal retinal angiogenesis.

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

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