

Ndst4 Cas9-CKO Strategy

Designer:

JiaYu

Reviewer:

Xiaojing Li

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Project Overview

Project Name

Ndst4

Project type

Cas9-CKO

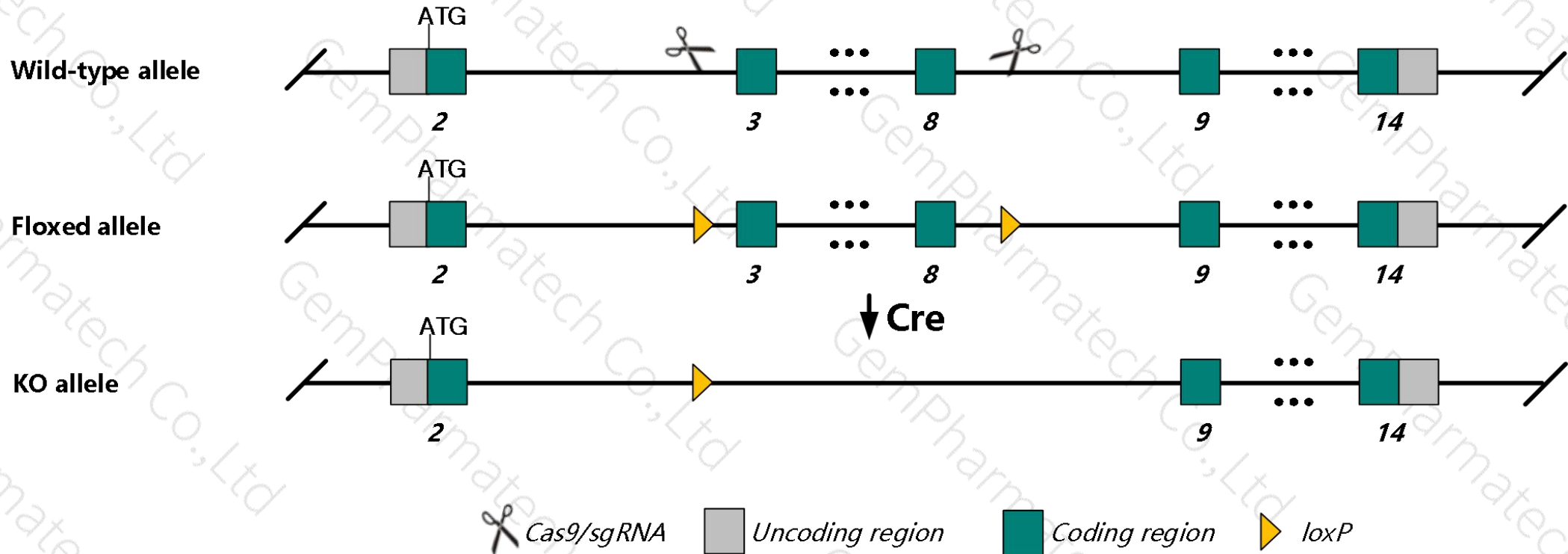
Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ndst4* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System



Technical routes

- The *Ndst4* gene has 6 transcripts. According to the structure of *Ndst4* gene, exon3-exon8 of *Ndst4-204* (ENSMUST00000173932.7) transcript is recommended as the knockout region. The region contains 838bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ndst4* 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 a phenotype restricted to the colonic epithelium that includes an increased number of colon goblet cells, a decreased number of colonocytes, and increased apoptosis of colonic epithelial cells in the proximal colon.
- Transcript 206 CDS 5' and 3' incomplete the influences is unknown.
- The *Ndst4* gene is located on the Chr3. 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)

Ndst4 N-deacetylase/N-sulfotransferase (heparin glucosaminyl) 4 [*Mus musculus* (house mouse)]

Gene ID: 64580, updated on 13-Mar-2020

Summary

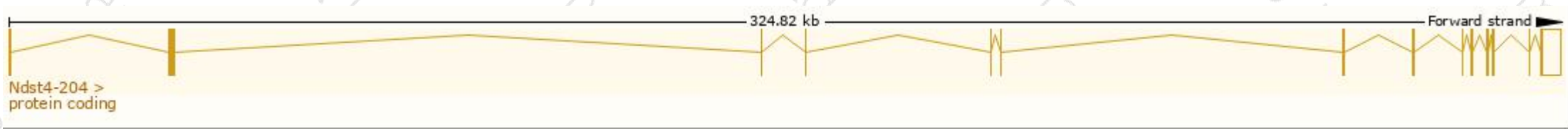
Official Symbol	Ndst4 provided by MGI
Official Full Name	N-deacetylase/N-sulfotransferase (heparin glucosaminyl) 4 provided by MGI
Primary source	MGI:MGI:1932545
See related	Ensembl:ENSMUSG00000027971
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	4930439H17Rik
Expression	Biased expression in frontal lobe adult (RPKM 2.2), testis adult (RPKM 1.9) and 7 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

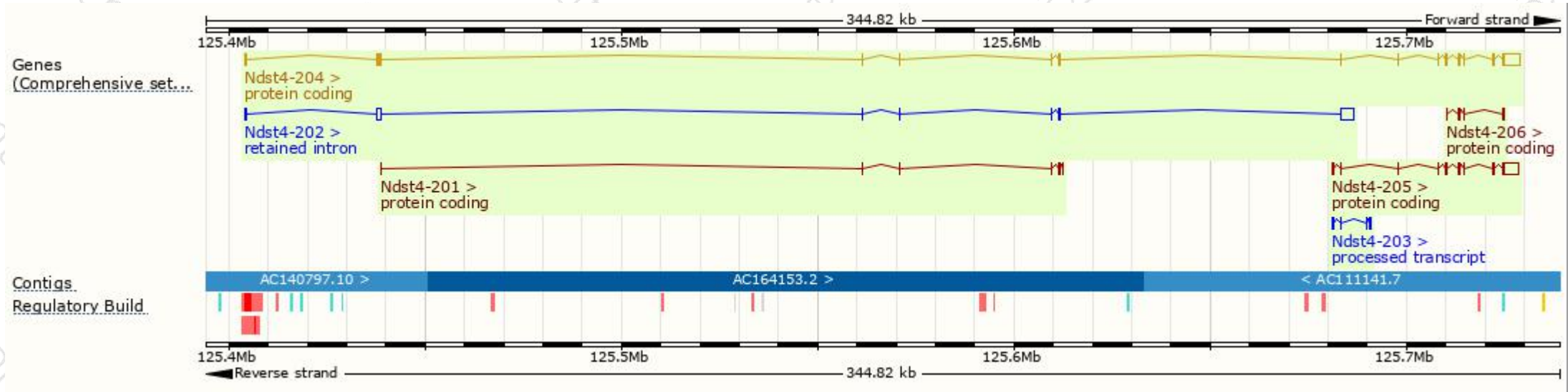
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ndst4-204	ENSMUST00000173932.7	7417	872aa	Protein coding	CCDS17820	Q9EQW8	TSL:5 GENCODE basic APPRIS P1
Ndst4-205	ENSMUST00000174648.5	4996	364aa	Protein coding	-	G3UX70	TSL:1 GENCODE basic
Ndst4-201	ENSMUST00000144344.2	718	205aa	Protein coding	-	D3YWZ6	CDS 5' incomplete TSL:5
Ndst4-206	ENSMUST00000198101.1	471	157aa	Protein coding	-	A0A0G2JDL4	CDS 5' and 3' incomplete TSL:3
Ndst4-203	ENSMUST00000172632.1	1042	No protein	Processed transcript	-	-	TSL:1
Ndst4-202	ENSMUST00000147016.4	5696	No protein	Retained intron	-	-	TSL:1

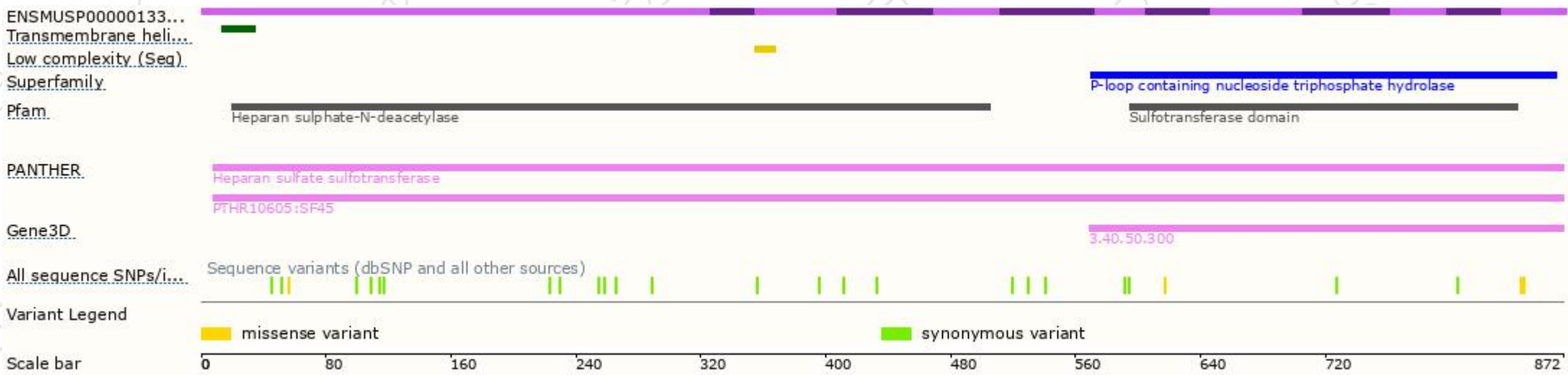
The strategy is based on the design of *Ndst4-204* transcript,The transcription is shown below



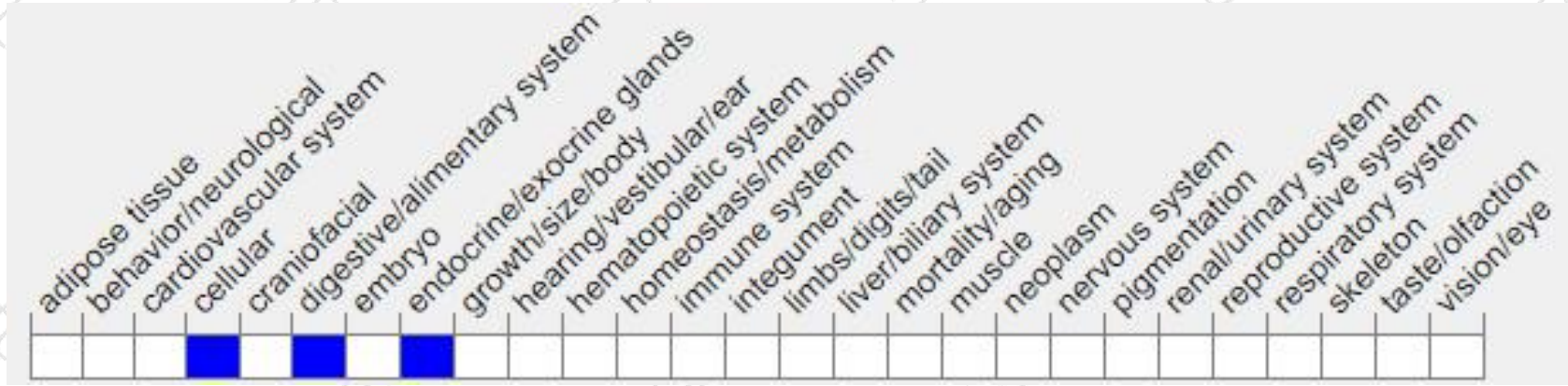
Genomic location distribution



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 a phenotype restricted to the colonic epithelium that includes an increased number of colon goblet cells, a decreased number of colonocytes, and increased apoptosis of colonic epithelial cells in the proximal colon.

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

Tel: 400-9660890

