

Hnf4a Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Hnf4a

Project type

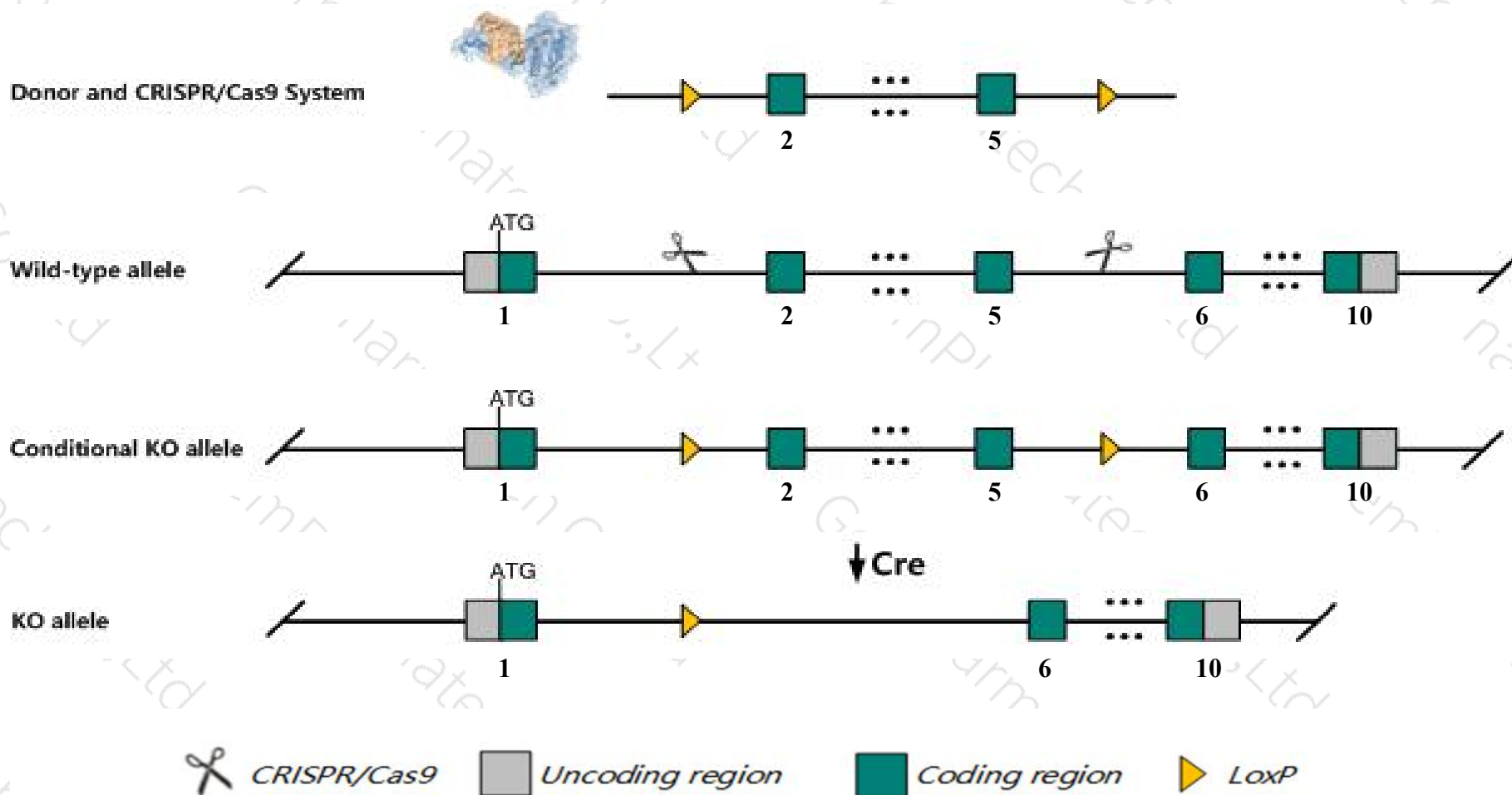
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hnf4a* gene. The schematic diagram is as follows:



Technical routes

- The *Hnf4a* gene has 5 transcripts. According to the structure of *Hnf4a* gene, exon2-exon5 of *Hnf4a*-201 (ENSMUST00000018094.12) transcript is recommended as the knockout region. The region contains 533bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Hnf4a* 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, Nullizygous embryos show delayed growth and lethality, impaired gastrulation, abnormal primitive streak and mesoderm formation, ectoderm apoptosis, and extraembryonic tissue dysplasia. Mice expressing only the alpha1 isoform show glucose intolerance whereas mice expressing alpha7 show dyslipidemia.
- The *Hnf4a* gene is located on the Chr2. 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)

Hnf4a hepatic nuclear factor 4, alpha [Mus musculus (house mouse)]

Gene ID: 15378, updated on 23-Mar-2019

Summary



Official Symbol	Hnf4a provided by MGI
Official Full Name	hepatic nuclear factor 4, alpha provided by MGI
Primary source	MGI:MGI:109128
See related	Ensembl:ENSMUSG00000017950
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	HNF-4, Hnf4, Hnf4alpha, MODY1, Nr2a1, TCF-14, Tcf14
Summary	The protein encoded by this gene is a transcription factor involved in the development of the pancreas, liver, kidney, and intestines. The encoded protein also functions to maintain glucose homeostasis. Several transcript variants encoding different isoforms have been found for this gene. [provided by RefSeq, Aug 2015]
Expression	Biased expression in large intestine adult (RPKM 148.8), kidney adult (RPKM 141.3) and 9 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

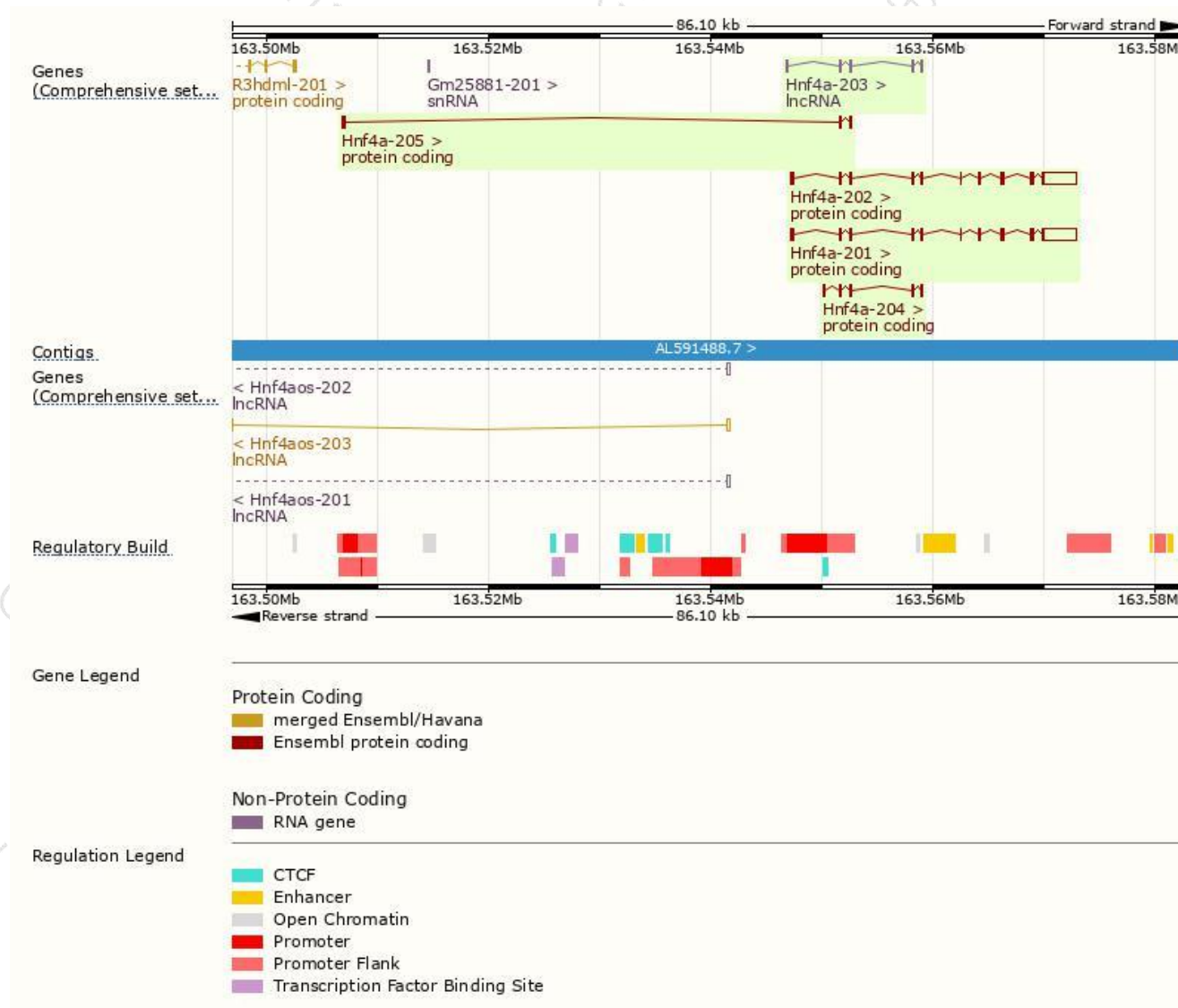
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hnf4a-201	ENSMUST00000018094.12	4356	474aa	Protein coding	CCDS17012	P49698	TSL:1 GENCODE basic APPRIS P1
Hnf4a-202	ENSMUST00000109411.7	4362	465aa	Protein coding	-	Z4YKX0	TSL:1 GENCODE basic
Hnf4a-204	ENSMUST00000137449.1	692	191aa	Protein coding	-	A2A5I6	CDS 3' incomplete TSL:3
Hnf4a-205	ENSMUST00000143911.7	431	100aa	Protein coding	-	A2A5I4	CDS 3' incomplete TSL:1
Hnf4a-203	ENSMUST00000131658.1	659	No protein	lncRNA	-	-	TSL:3

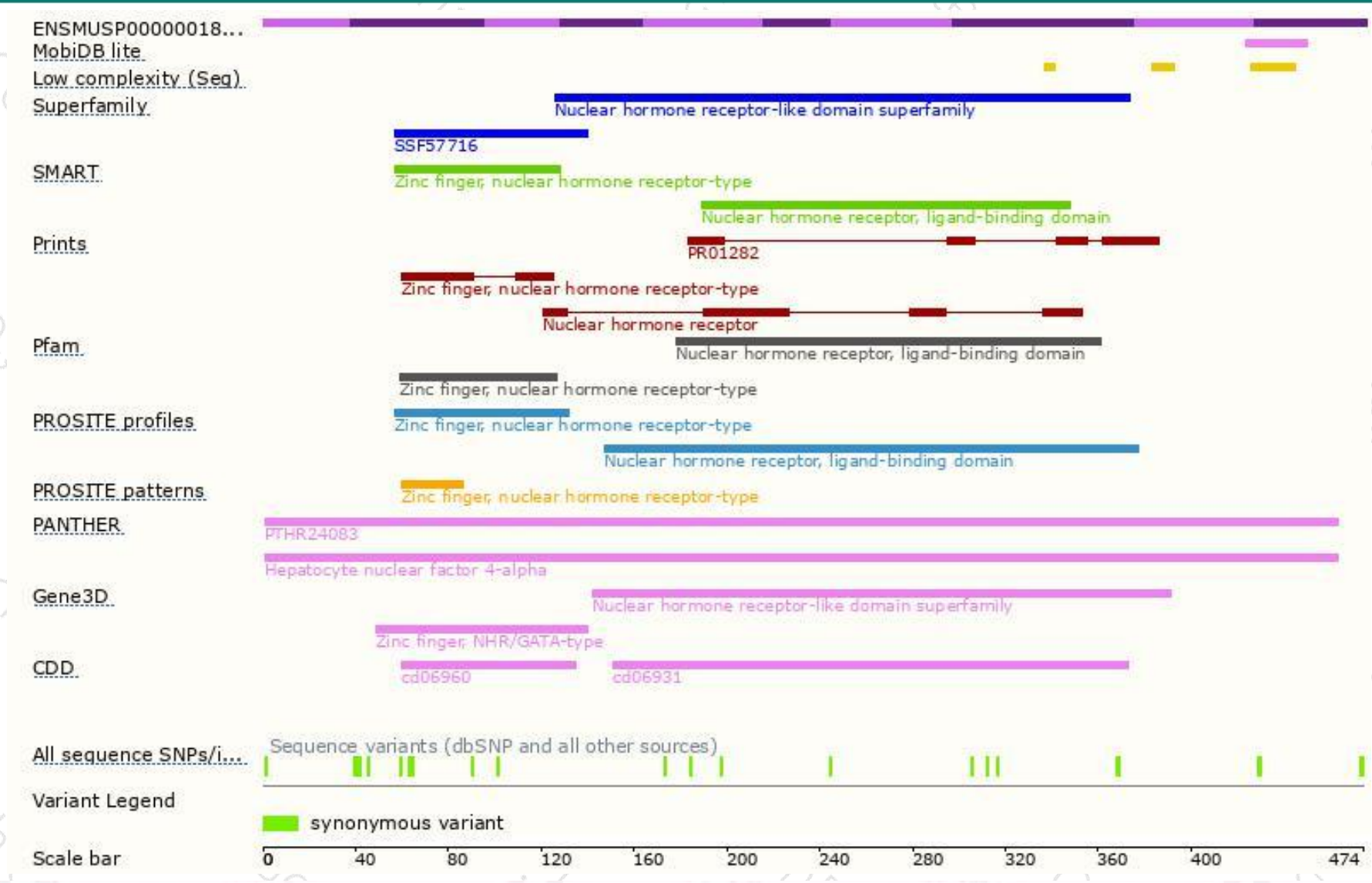
The strategy is based on the design of *Hnf4a-201* transcript,The transcription is shown below



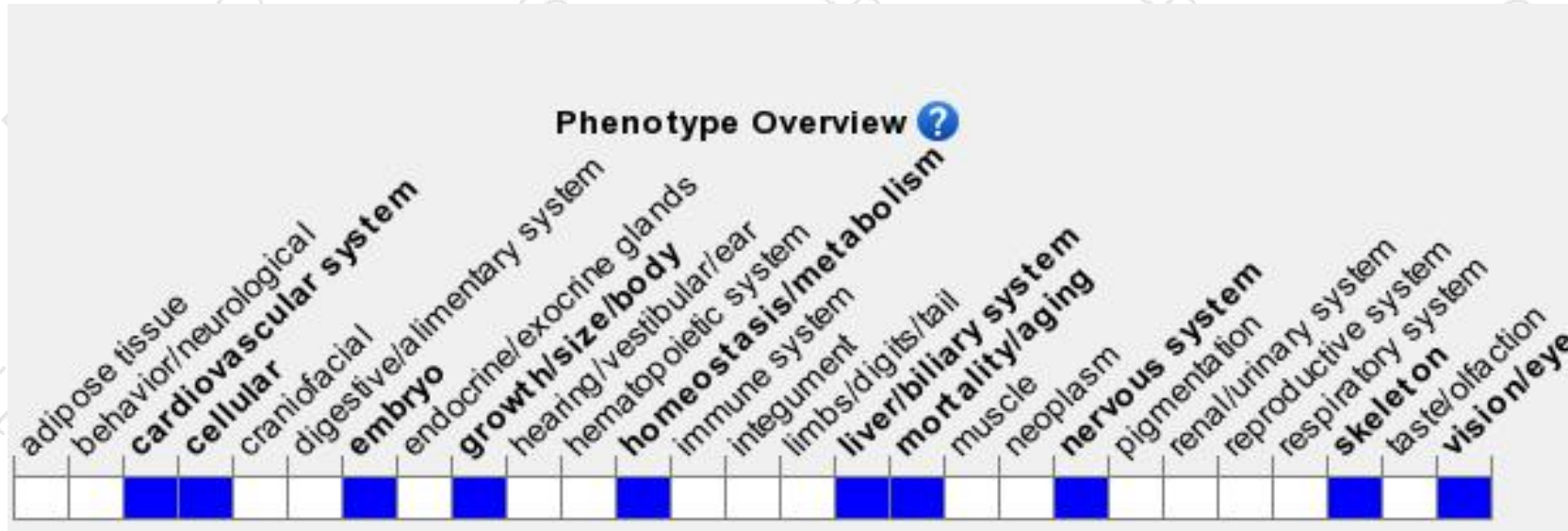
Genomic location distribution



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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