

Hoxd1 Cas9-KO Strategy

Designer:

Daohua Xu

Reviewer :

Huimin Su

Design Date:

2020-2-19

Project Overview

Project Name

Hoxd1

Project type

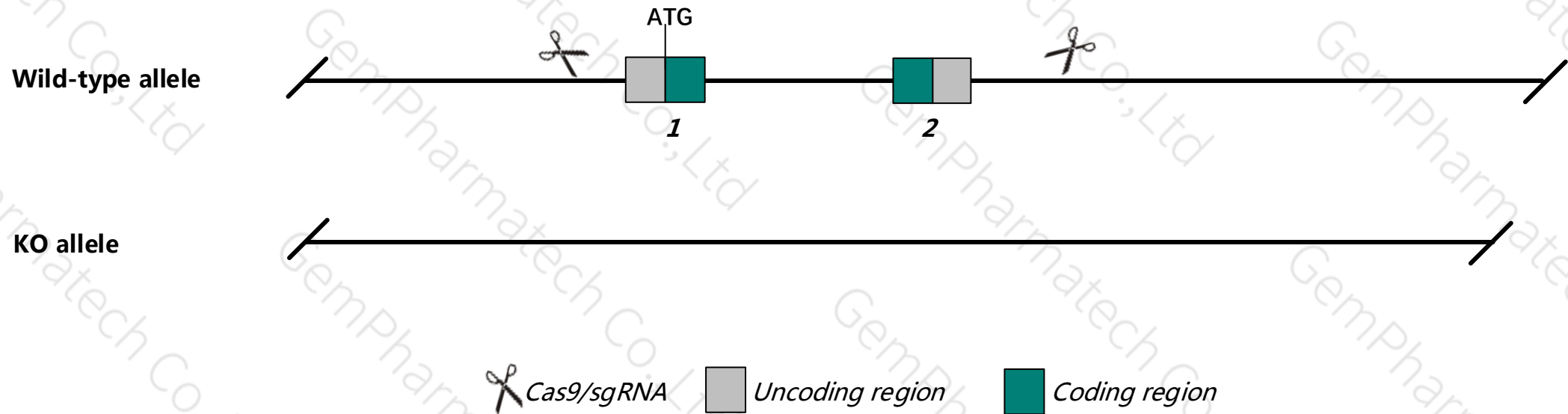
Cas9-KO

Strain background

C57BL/6JGpt

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hoxd1* gene. The schematic diagram is as follows:



Technical routes

- The *Hoxd1* gene has 1 transcript. According to the structure of *Hoxd1* gene, exon1-exon2 of *Hoxd1*-201 (ENSMUST00000047793.5) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Hoxd1* gene. The brief process is as follows: sgRNA was transcribed in vitro. Cas9, sgRNA 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.

- According to the existing MGI data , Mice homozygous for a reporter allele display abnormal cervical vertebrae. Mice homozygous for a knock-out allele exhibit abnormal nociceptor innervation of the skin, aberrant nociceptor axonal projections in the spinal cord, and deficits in pain sensitivity.
- The KO region contains functional region of the *Haglr* gene. Knockout the region may affect the function of *Haglr* gene.
- The *Hoxd1* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Hoxd1 homeobox D1 [*Mus musculus* (house mouse)]

Gene ID: 15429, updated on 12-Aug-2019

Summary

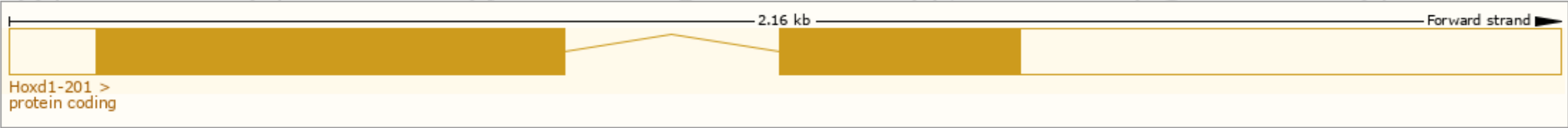
Official Symbol	Hoxd1 provided by MGI
Official Full Name	homeobox D1 provided by MGI
Primary source	MGI:MGI:96201
See related	Ensembl:ENSMUSG00000042448
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Hox-4.9
Expression	Low expression observed in reference dataset See more
Orthologs	human all

Transcript information (Ensembl)

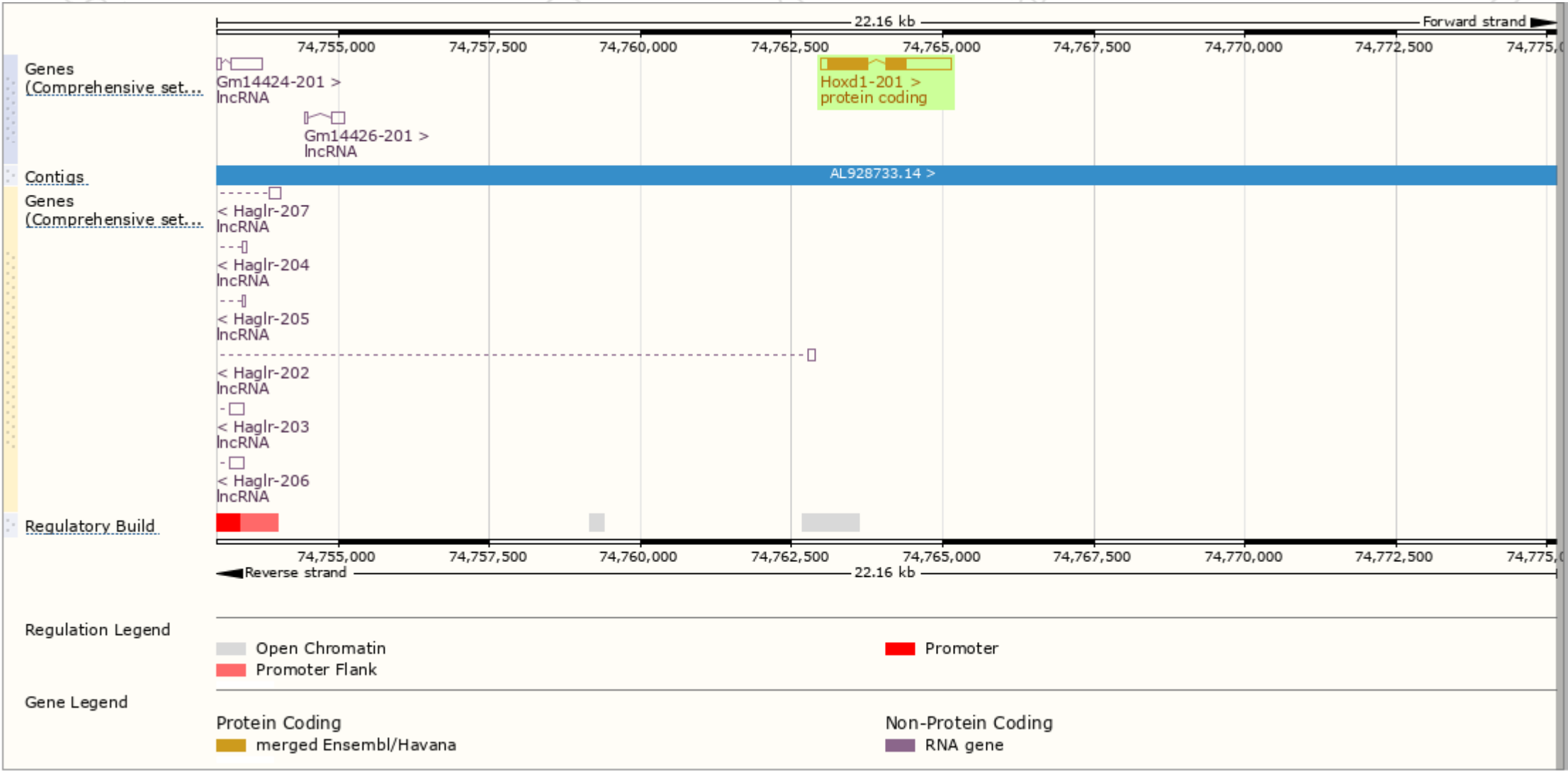
The gene has 1 transcript, and the transcripts is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hoxd1-201	ENSMUST00000047793.5	1862	328aa	Protein coding	CCDS16147	Q01822	TSL:1 GENCODE basic APPRIS P1

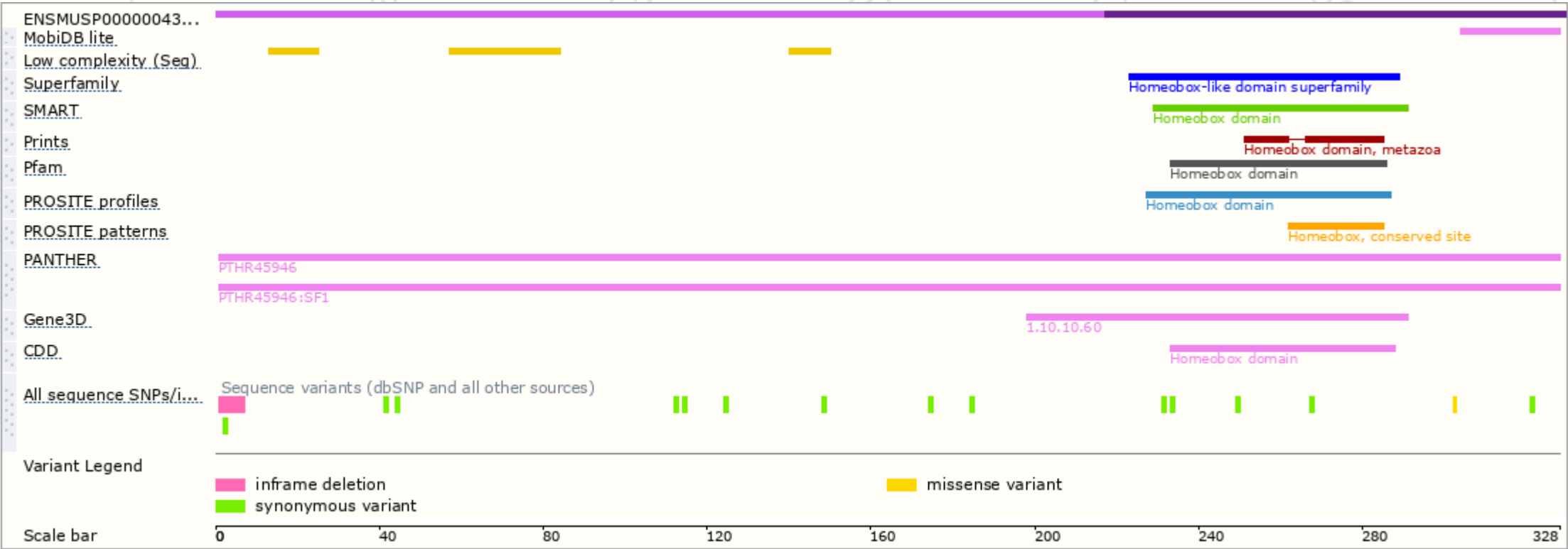
The strategy is based on the design of *Hoxd1*-201 transcript,The transcription is shown below



Genomic location distribution

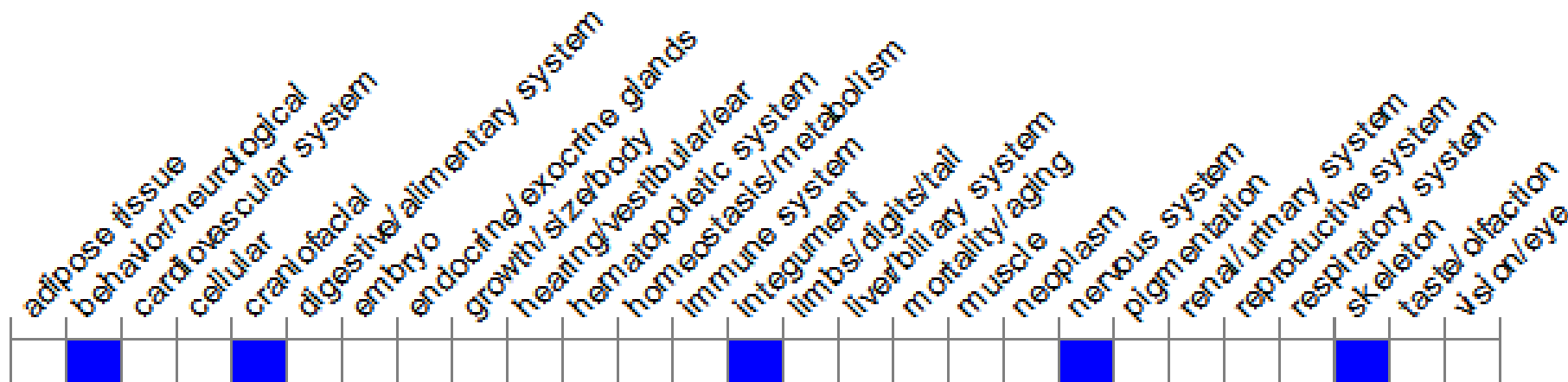


Protein domain



Mouse phenotype description(MGI)

Phenotype Overview ?



Click cells to view annotations.

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 reporter allele display abnormal cervical vertebrae.

Mice homozygous for a knock-out allele exhibit abnormal nociceptor innervation of the skin, aberrant nociceptor axonal projections in the spinal cord, and deficits in pain sensitivity.

If you have any questions, you are welcome to inquire.
Tel: 025-5864 1534

