

Dhh Cas9-CKO Strategy

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Project Overview

Project Name

Dhh

Project type

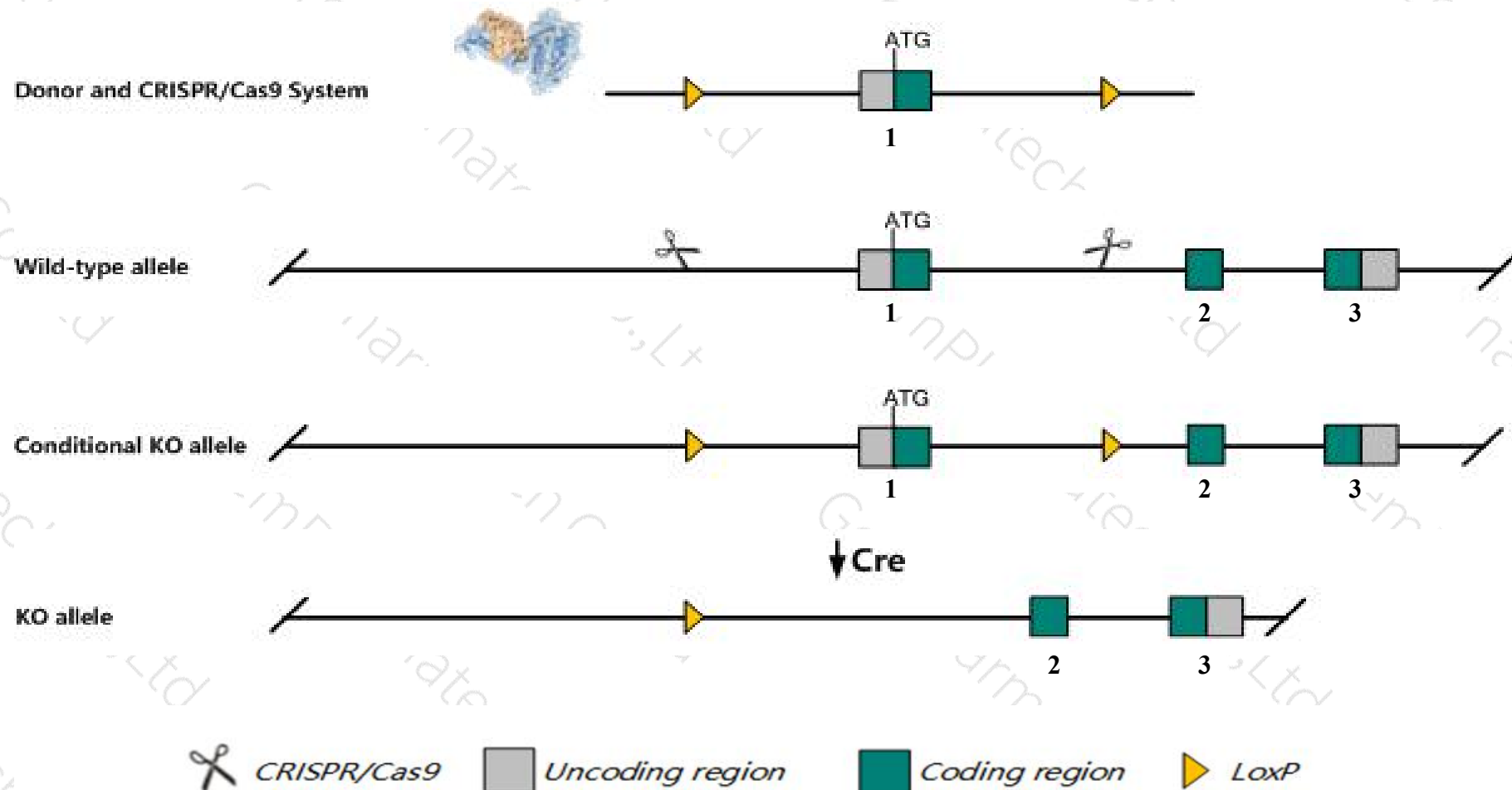
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- The *Dhh* gene has 2 transcripts. According to the structure of *Dhh* gene, exon1 of *Dhh-201* (ENSMUST00000023737.5) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Dhh* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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, Homozygous null mutants are male sterile, failing to produce mature spermatozoa; peripheral nerves are abnormal, with thin and disorganized perineurial sheaths. High penetrance of pseudohermaphroditism observed on some mixed backgrounds.
- The insertion of 3-terminal loxp may affect the activation of the *Rheb11* gene
- The *Dhh* gene is located on the Chr15. 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)

Dhh desert hedgehog [Mus musculus (house mouse)]

Gene ID: 13363, updated on 9-Apr-2019

Summary



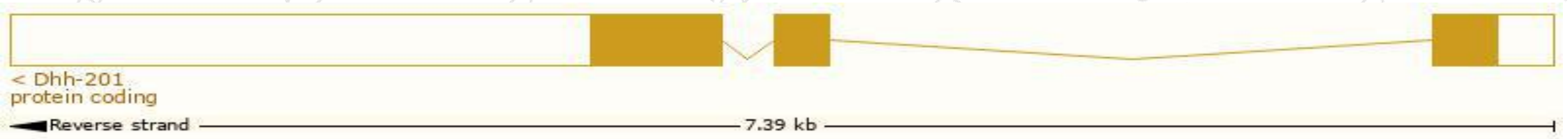
Official Symbol	Dhh provided by MGI
Official Full Name	desert hedgehog provided by MGI
Primary source	MGI:MGI:94891
See related	Ensembl:ENSMUSG00000023000
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	C78960
Expression	Biased expression in ovary adult (RPKM 10.4), testis adult (RPKM 4.0) and 7 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

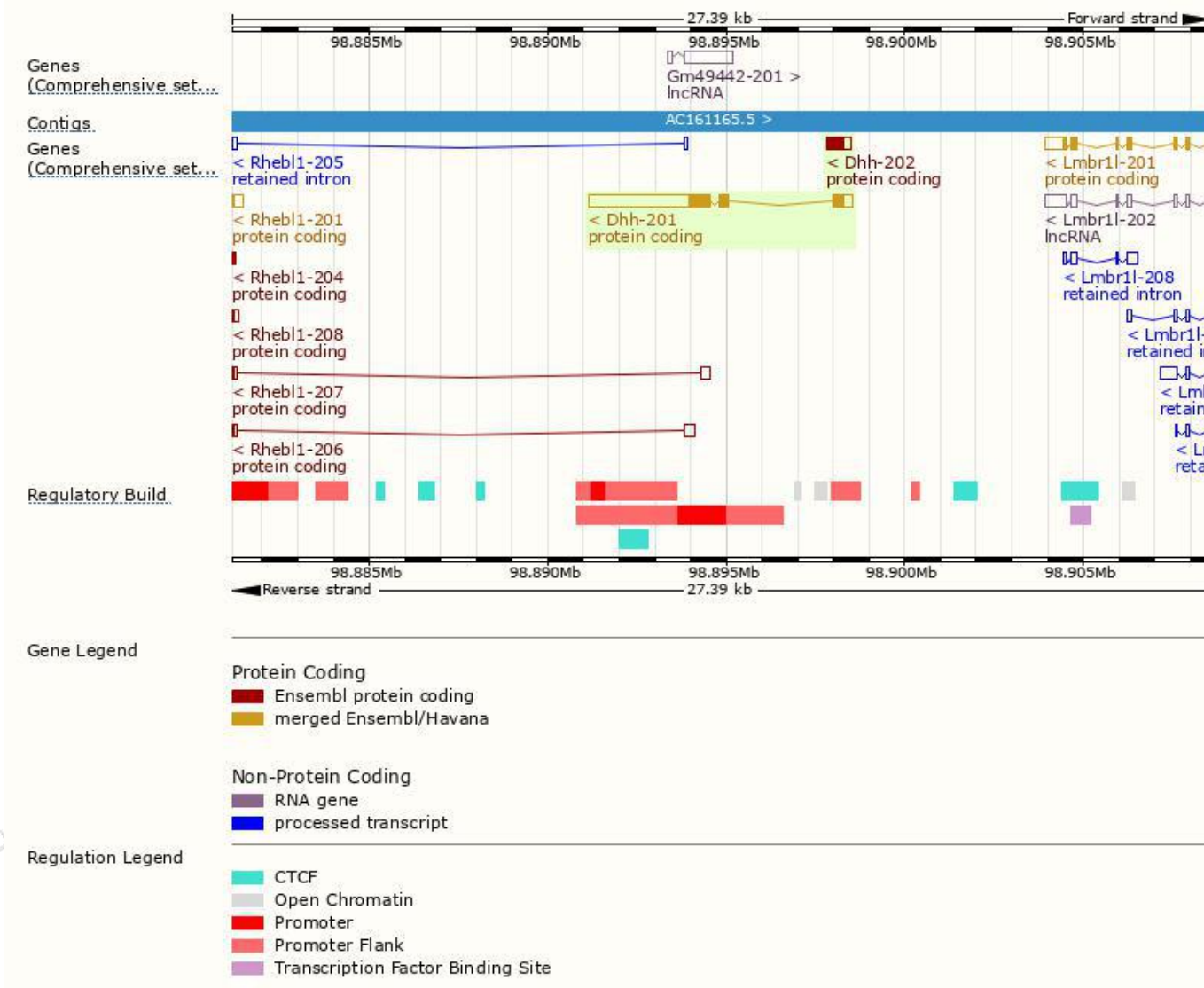
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dhh-201	ENSMUST00000023737.5	4242	396aa	Protein coding	CCDS27810	Q544P6 Q61488	TSL:1 GENCODE basic APPRIS P1
Dhh-202	ENSMUST00000229775.1	694	138aa	Protein coding	-	A0A2R8VI43	GENCODE basic

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



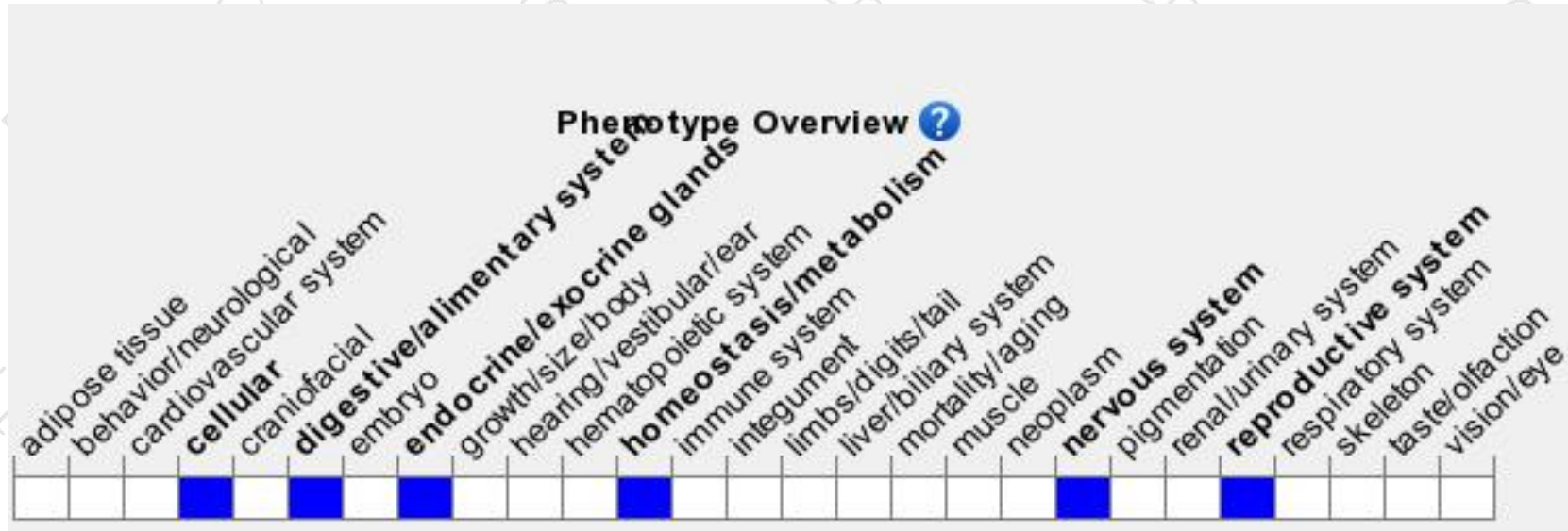
Genomic location distribution



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, Homozygous null mutants are male sterile, failing to produce mature spermatozoa; peripheral nerves are abnormal, with thin and disorganized perineurial sheaths. High penetrance of pseudohermaphroditism observed on some mixed backgrounds.

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

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