



# ***Hoxd11 Cas9-CKO Strategy***

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# Project Overview

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**Project Name**

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***Hoxd11***

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**Project type**

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**Cas9-CKO**

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**Strain background**

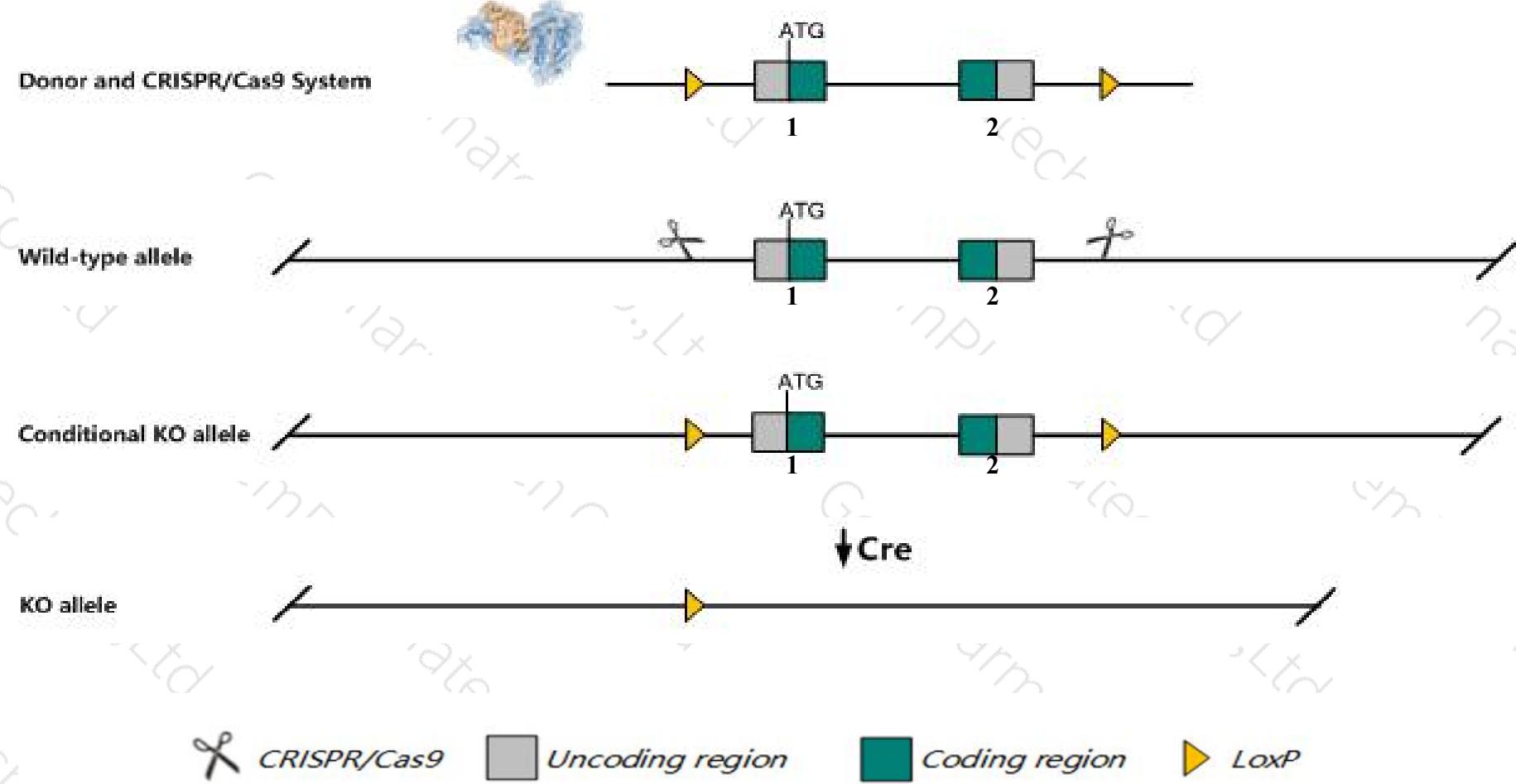
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**C57BL/6JGpt**

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# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hoxd11* gene. The schematic diagram is as follows:



# Technical routes

- The *Hoxd11* gene has 2 transcripts. According to the structure of *Hoxd11* gene, exon1-exon2 of *Hoxd11-202* (NSMUST00000142312.2) 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 *Hoxd11* 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.



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# Notice

- According to the existing MGI data, Homozygotes for targeted null mutations exhibit homeotic transformations of sacral vertebrae, malformations of distal limbs, and reduced fertility in males.
- *Gm28309* gene will be deleted together.
- The floxed region is near to the N-terminal of *Hoxd10* gene and C-terminal of *Hoxd12* gene, this strategy may influence the regulatory function of the N-terminal of *Hoxd10* gene and C-terminal of *Hoxd12* gene.
- The *Hoxd11* 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)

## Hoxd11 homeobox D11 [ *Mus musculus* (house mouse) ]

Gene ID: 15431, updated on 10-Mar-2020

### Summary

Official Symbol	Hoxd11 provided by MGI
Official Full Name	homeobox D11 provided by MGI
Primary source	MGI:MGI:96203
See related	<a href="#">Ensembl:ENSMUSG00000042499</a>
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Hox-4.6; Hox-5.4; Hox-5.5; E230017H14Rik
Expression	Biased expression in limb E14.5 (RPKM 8.5), subcutaneous fat pad adult (RPKM 4.6) and 5 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

### Genomic context

Location: 2 C3; 2 44.13 cM

See Hoxd11 in [Genome Data Viewer](#)

Exon count: 3

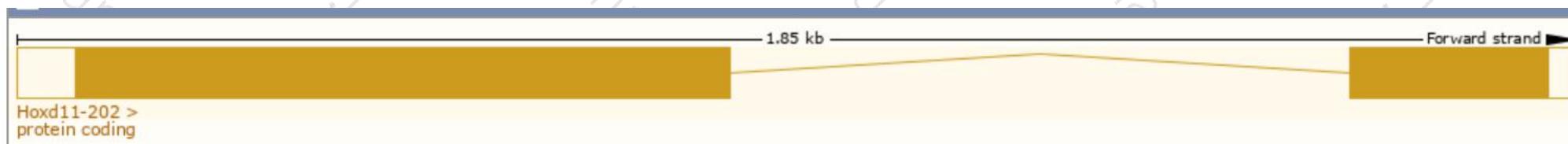
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 ( <a href="#">GCF_000001635.26</a> )	2	NC_000068.7 (74679558..74687016)
Build 37.2	previous assembly	MGSCv37 ( <a href="#">GCF_000001635.18</a> )	2	NC_000068.6 (74520450..74522195)

# Transcript information (Ensembl)

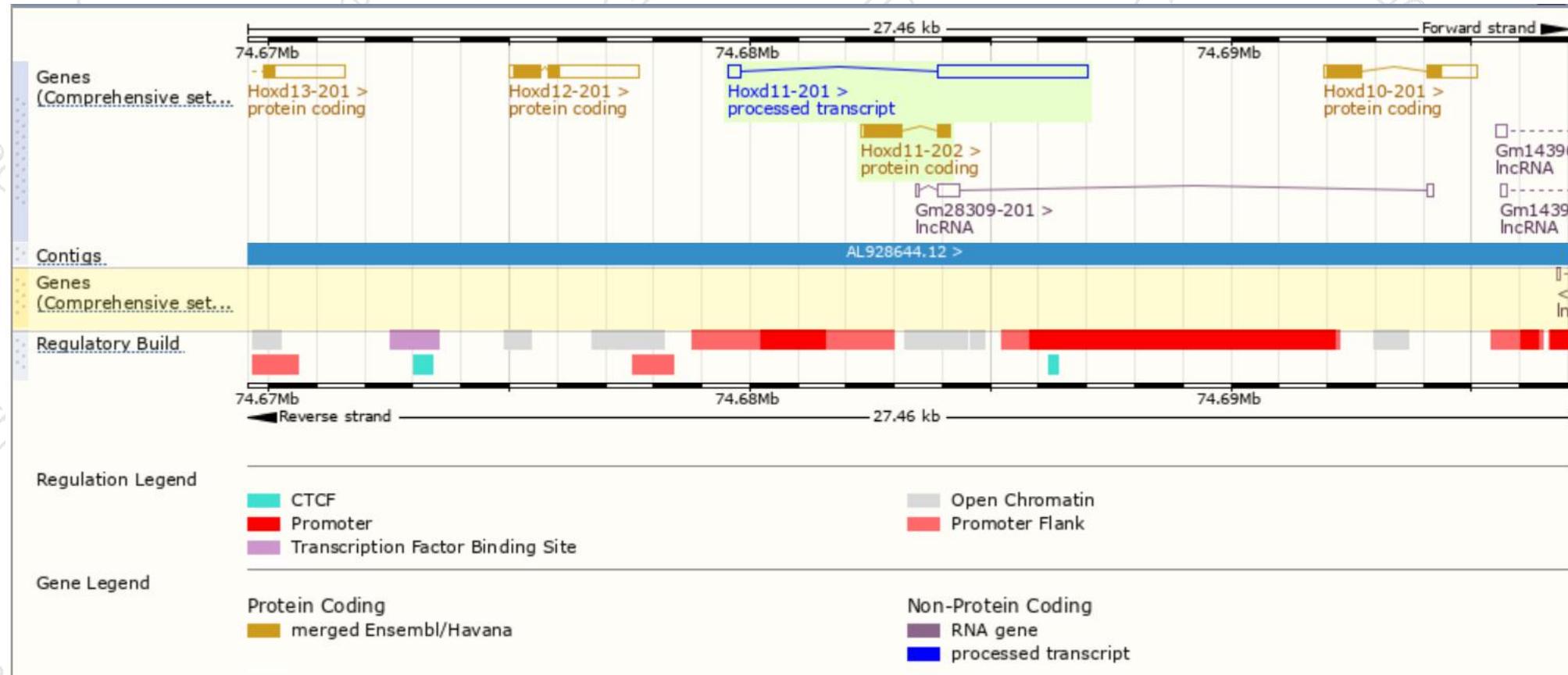
The gene has 2 transcripts, and all the transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hoxd11-202	<a href="#">ENSMUST00000142312.2</a>	1111	336aa	Protein coding	<a href="#">CCDS16140</a>	<a href="#">A2ASM7</a>	TSL:1 GENCODE basic APPRIS P1
Hoxd11-201	<a href="#">ENSMUST0000048086.8</a>	3367	No protein	Processed transcript	-	-	TSL:1

The strategy is based on the design of *Hoxd11-202* transcript, The transcription is shown below



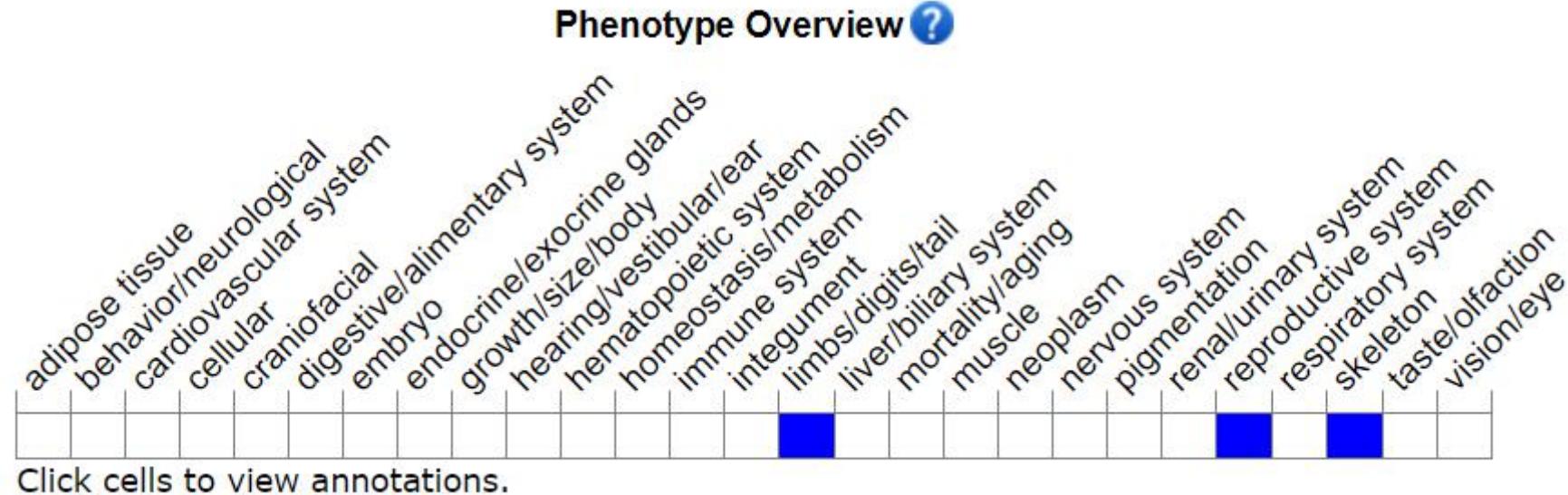
# Genomic location distribution



# 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/>).*

Homozygotes for targeted null mutations exhibit homeotic transformations of sacral vertebrae, malformations of distal limbs, and reduced fertility in males.



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

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