

# ***E2f4 Cas9-KO Strategy***

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# Project Overview

**Project Name**

***E2f4***

**Project type**

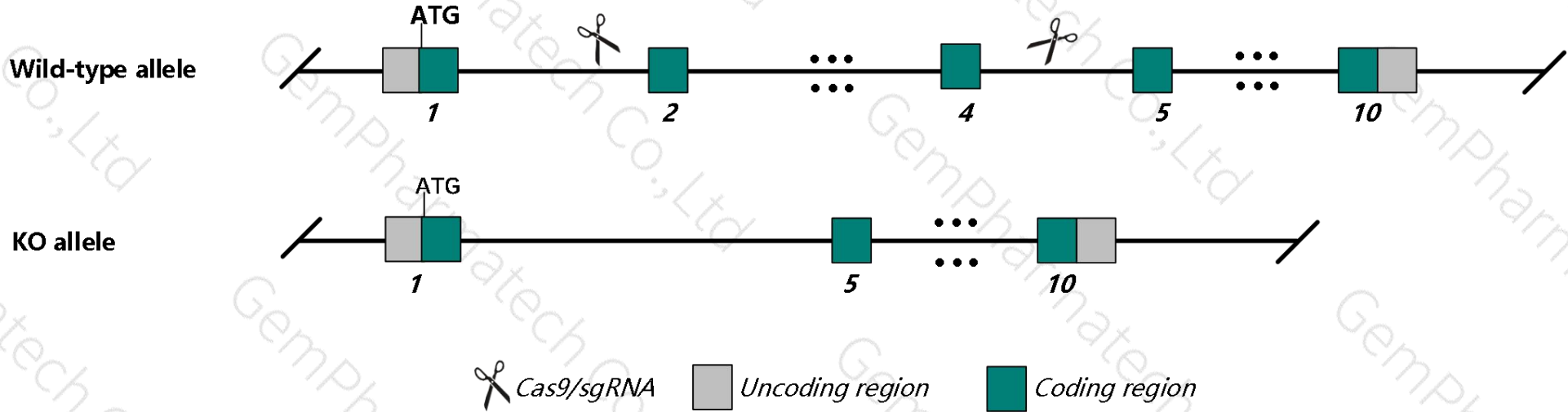
**Cas9-KO**

**Strain background**

**C57BL/6J**

# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *E2f4* gene. The schematic diagram is as follows:



# Technical routes

- The *E2f4* gene has 7 transcripts. According to the structure of *E2f4* gene, exon2-4 of *E2f4-201* (ENSMUST00000015003.9) transcript is recommended as the knockout region. The region contains 316bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *E2f4* gene. The brief process is as follows: sgRNA was transcribed in vitro. Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J mice. Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. The stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6J mice.

- According to the existing MGI data, homozygous null mice die postnatally of an increased susceptibility to bacterial infection and exhibit craniofacial defects, erythroid abnormalities, and growth retardation.
- The *E2f4* gene is located on the Chr8. 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.
- There are two coding genes near *E2f4* gene ( *Exoc31* and *Elmo3* ), and the deletion of *E2f4* gene may affect the expression of two adjacent genes.
- 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)

## E2f4 E2F transcription factor 4 [ *Mus musculus* (house mouse) ]

Gene ID: 104394, updated on 11-Jun-2019

### Summary

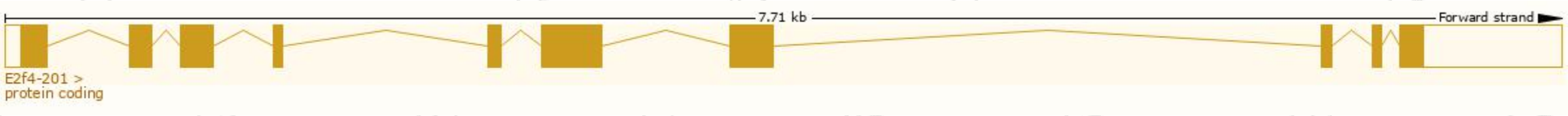
Official Symbol	E2f4 provided by <a href="#">MGI</a>
Official Full Name	E2F transcription factor 4 provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:103012</a>
See related	<a href="#">Ensembl:ENSMUSG00000014859</a>
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI427446; 2010111M04Rik
Expression	Ubiquitous expression in liver E14.5 (RPKM 188.0), liver E14 (RPKM 147.2) and 28 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

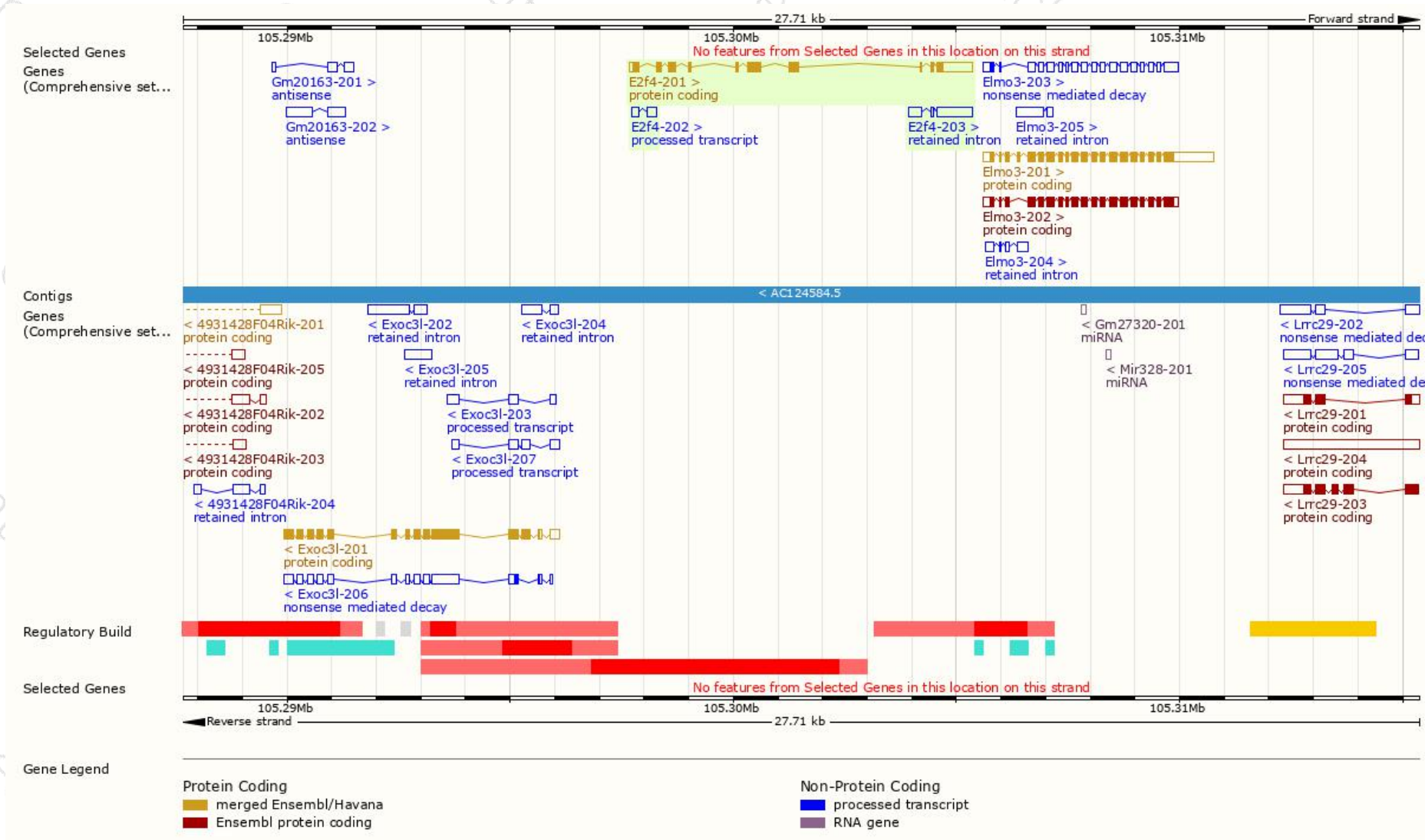
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
E2f4-201	<a href="#">ENSMUST00000015003.9</a>	1993	<a href="#">410aa</a>	Protein coding	<a href="#">CCDS40456</a>	<a href="#">Q8R0K9</a>	TSL:1 GENCODE basic APPRIS P1
E2f4-202	<a href="#">ENSMUST000000212037.1</a>	358	No protein	Processed transcript	-	-	TSL:3
E2f4-203	<a href="#">ENSMUST000000212572.1</a>	1151	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *E2f4-201* transcript, the transcription is shown below:



# Genomic location distribution

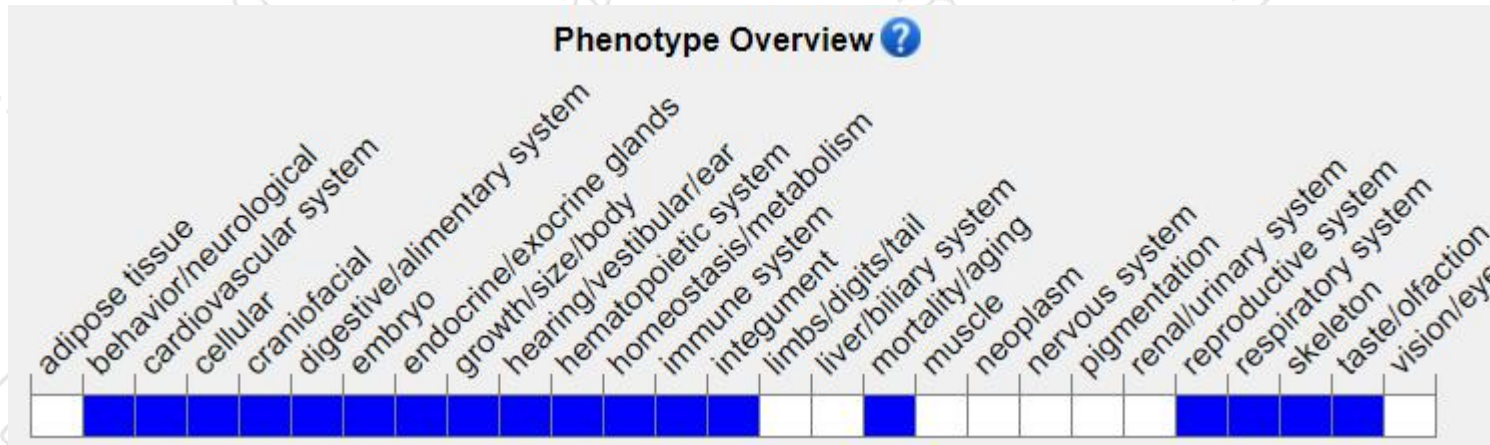




# Protein domain



# Mouse phenotype description(MGI )



*Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).*

Homozygous null mice die postnatally of an increased susceptibility to bacterial infection and exhibit craniofacial defects, erythroid abnormalities, and growth retardation.

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

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