Atf4 Cas9-KO *Strategy*

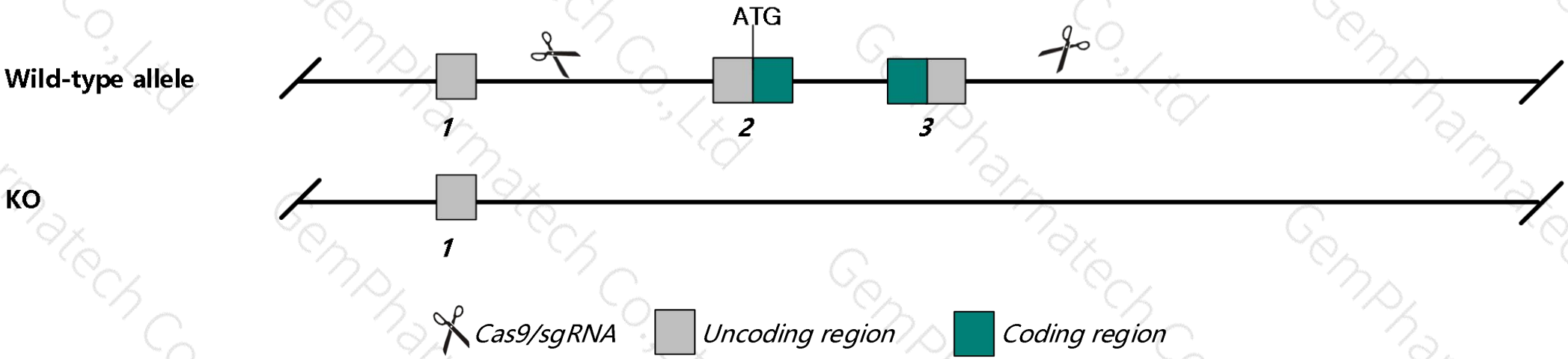
Designer: **Daohua Xu**

Project Overview

Project Name	***Atf4***
Project type	**Cas9-KO**
Strain background	**C57BL/6J**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Atf4* gene. The schematic diagram is as follows:



Technical routes

- The *Atf4* gene has 3 transcripts.According to the structure of *Atf4* gene, exon2-exon3 of *Atf4*-201 (ENSMUST00000109605.4)transcript is recommended as the knockout region.The region contains all coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Atf4* 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. A stable F1 generation mouse model was obtained by mating Positive F0 generation mice with C57BL/6J mice.

Notice

- According to the existing MGI data，Mice homozygous for one knock-out allele exhibit postnatal lethality, abnormal lens development, and reduced male fertility. Mice homozygous for a different knock-out allele exhibit abnormal pancreatic and skeletal development, glucose homeostasis, and insulin homeostasis.
- The *Atf4* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information （NCBI)

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Atf4 activating transcription factor 4 [*Mus musculus* (house mouse)]

Gene ID: 11911, updated on 30-Apr-2019

Summary

Official Symbol	Atf4 provided by MGI
Official Full Name	activating transcription factor 4 provided by MGI
Primary source	MGI:MGI:88096
See related	Ensembl:ENSMUSG00000042406
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	Atf-4; C/ATF; CREB2; TAXREB67
Expression	Ubiquitous expression in liver E14 (RPKM 153.4), liver E14.5 (RPKM 138.1) and 28 other tissues See more
Orthologs	human all

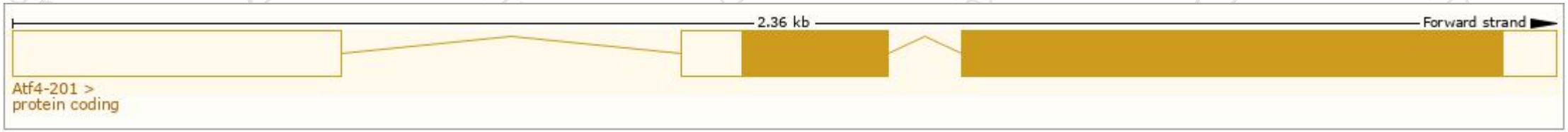
Transcript information （Ensembl）

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

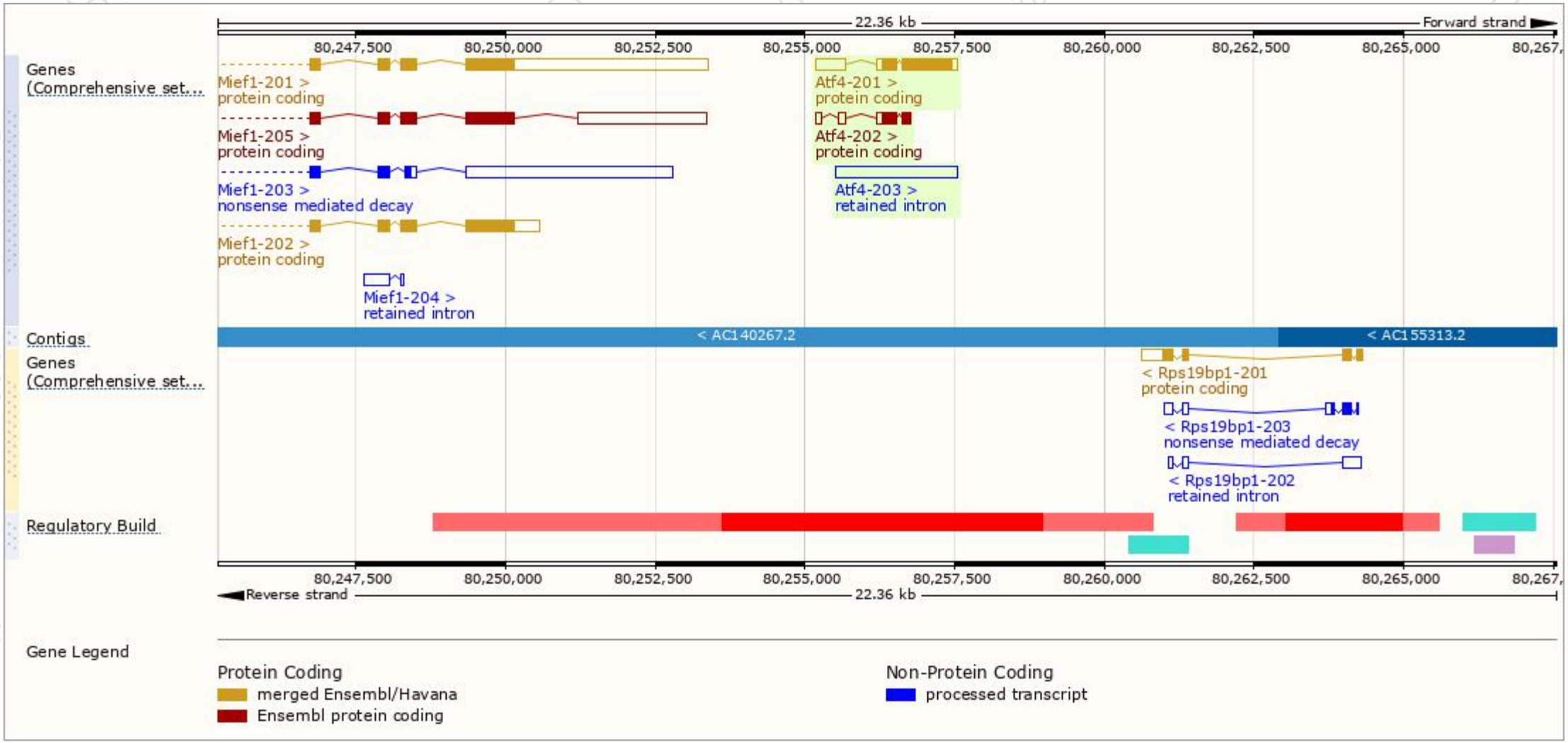
Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atf4-201	ENSMUST00000109605.4	1725	349aa	Protein coding	CCDS37145	Q06507	TSL:1 GENCODE basic APPRIS P1
Atf4-202	ENSMUST00000229828.1	659	116aa	Protein coding	-	A0A2R8VI82	CDS 3' incomplete
Atf4-203	ENSMUST00000230434.1	2019	No protein	Retained intron	-	-	-

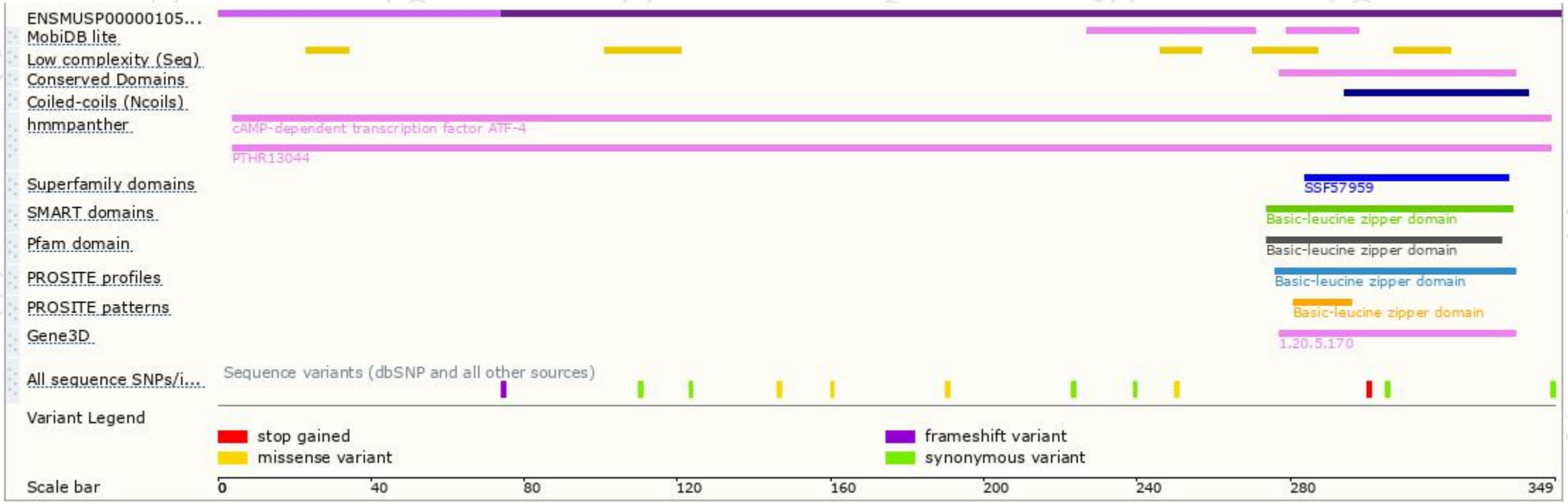
The strategy is based on the design of *Atf4-201* transcript,The transcription is shown below



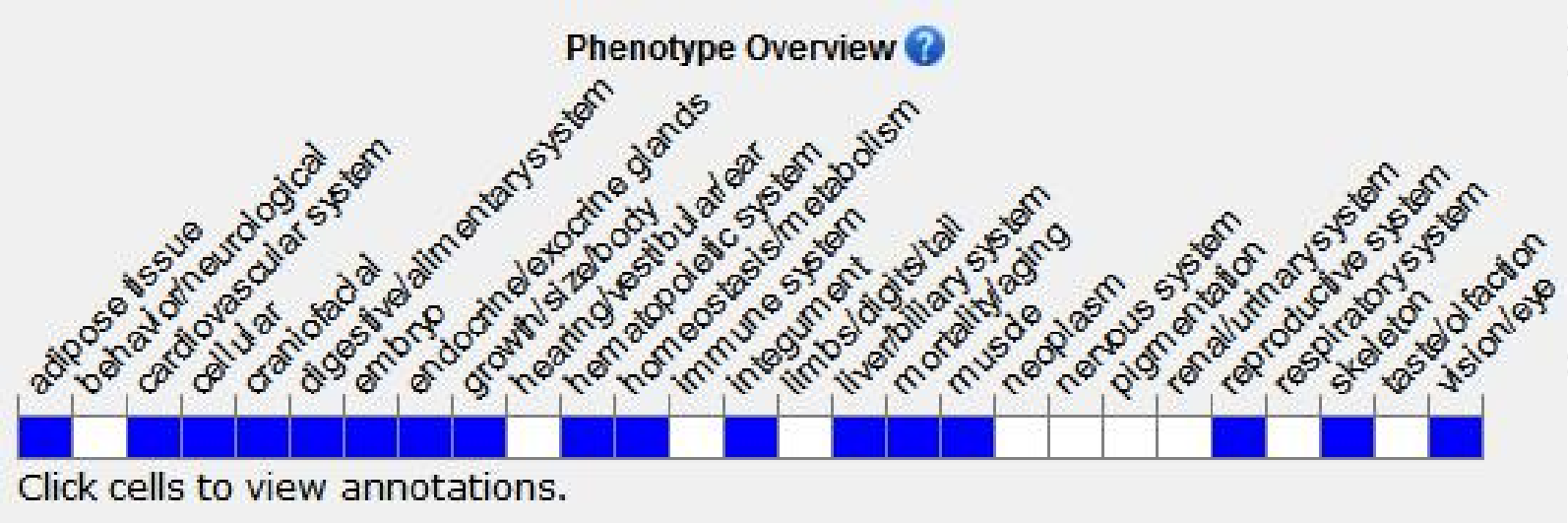
Genomic location (Ensembl)



Protein domain (Ensembl)



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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