Myo1e Cas9-KO Strategy

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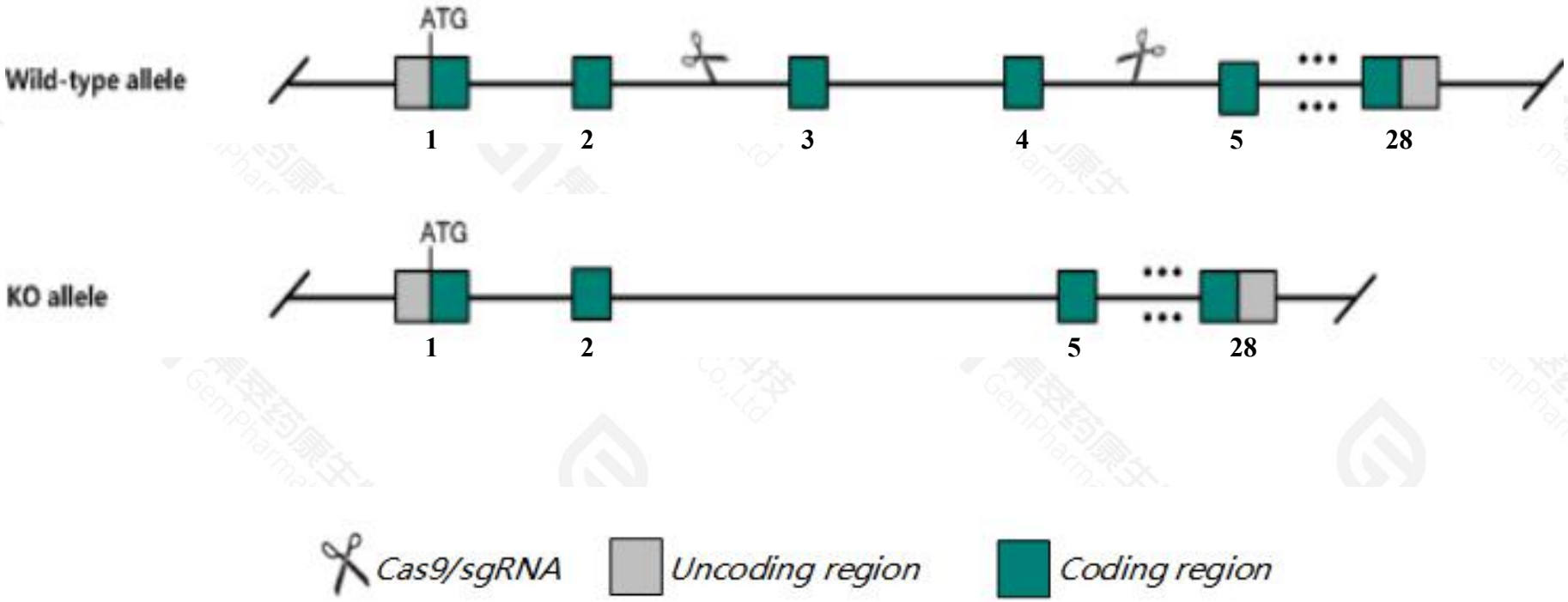
Design Date: 2021-5-31

Project Overview

Project Name	***Myo1e***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Myo1e* gene. The schematic diagram is as follows:



Technical routes

➢ The *Myo1e* gene has 4 transcripts. According to the structure of *Myo1e* gene, exon3-exon4 of *Myo1e-201*(ENSMUST00000034745.9) transcript is recommended as the knockout region.The region contains 185bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Myo1e* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and 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.

Notice

➢ According to the existing MGI data,homozygotes for a gene trapped allele exhibit embryonic lethality, embryonic hemorrhaging and hematopoietic defects. Homozygotes for a knock-out allele show proteinuria, chronic renal injury, kidney inflammation, and defects in renal filtration and podocyte organization.

➢ The *Myo1e* gene is located on the Chr9. 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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Myo1e myosin IE [Mus musculus (house mouse)]

Gene ID: 71602, updated on 26-Jan-2021

Summary

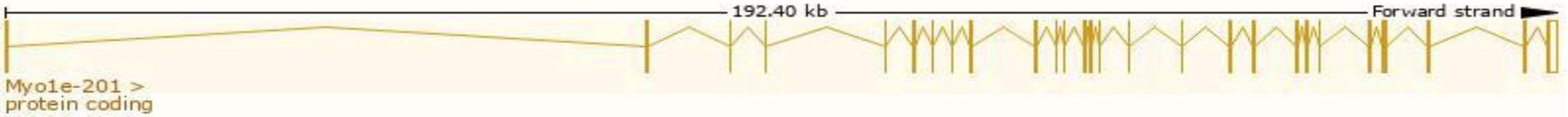
Official Symbol	Myo1e provided by MGI
Official Full Name	myosin IE provided byMGI
Primary source	MGI:MGI:106621
See related	Ensembl:ENSMUSG00000032220
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2310020N23Rik, 9130023P14Rik, AA407778, myosin-1e, myr 3
Expression	Ubiquitous expression in colon adult (RPKM 9.1), lung adult (RPKM 7.2) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

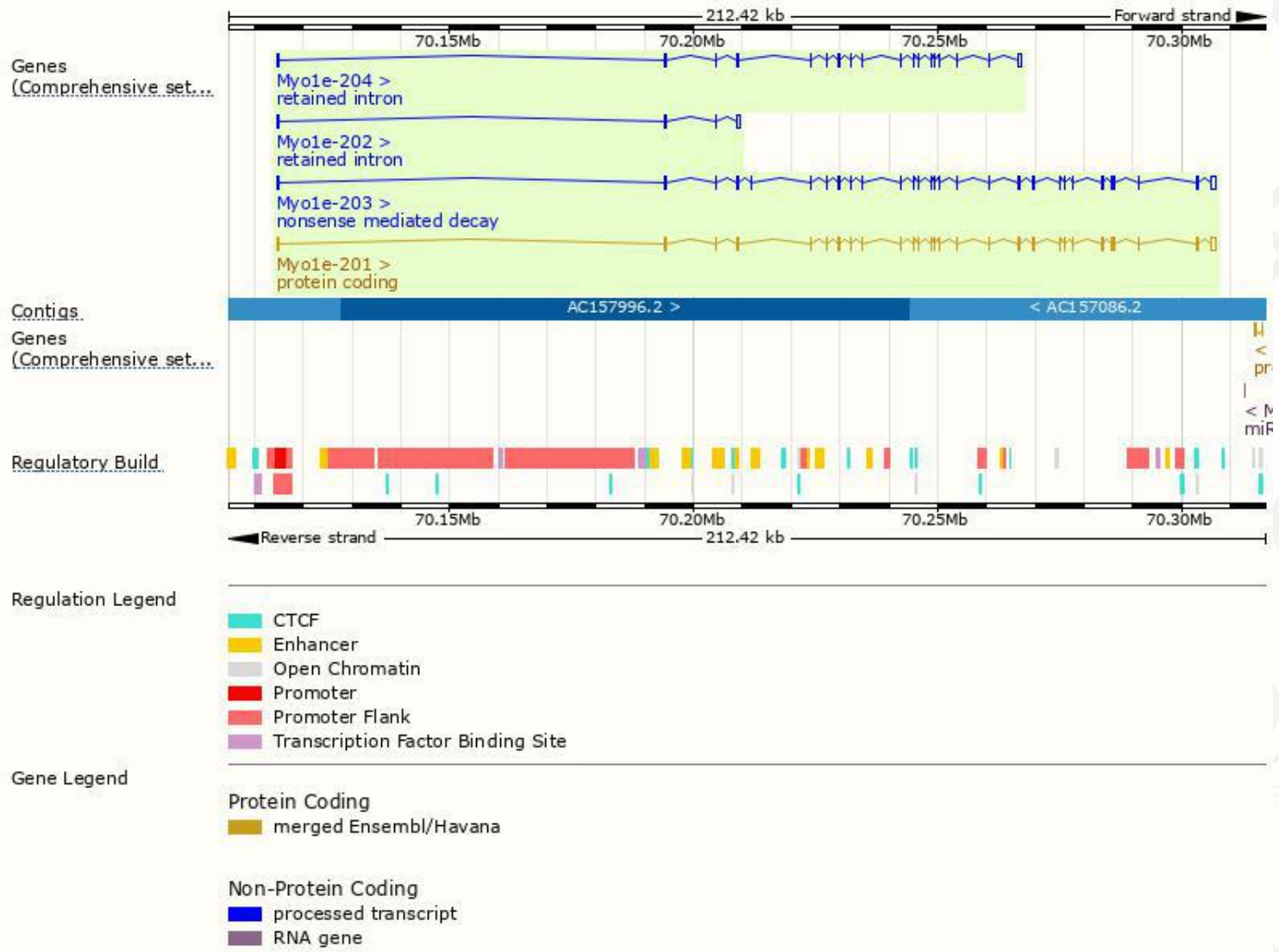
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Myo1e-201	ENSMUST00000034745.9	4625	1107aa	Protein coding	CCDS40680		TSL:1 , GENCODE basic , APPRIS P1 ,
Myo1e-203	ENSMUST00000214042.2	4699	116aa	Nonsense mediated decay	-		TSL:1 ,
Myo1e-204	ENSMUST00000214767.2	2671	No protein	Retained intron	-		TSL:1 ,
Myo1e-202	ENSMUST00000213863.2	1260	No protein	Retained intron	-		TSL:1 ,

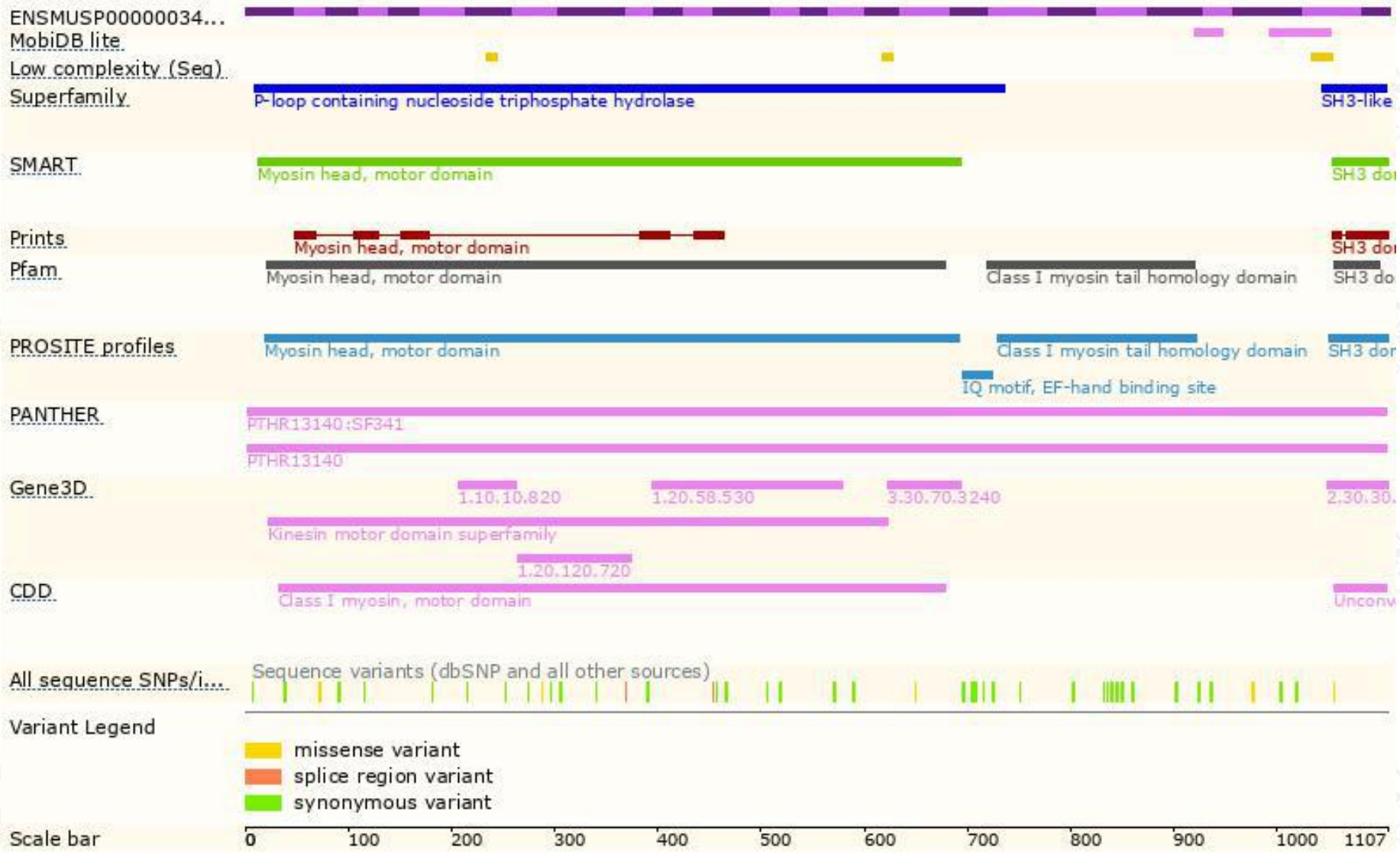
The strategy is based on the design of *Myo1e-201* transcript,the transcription is shown below:



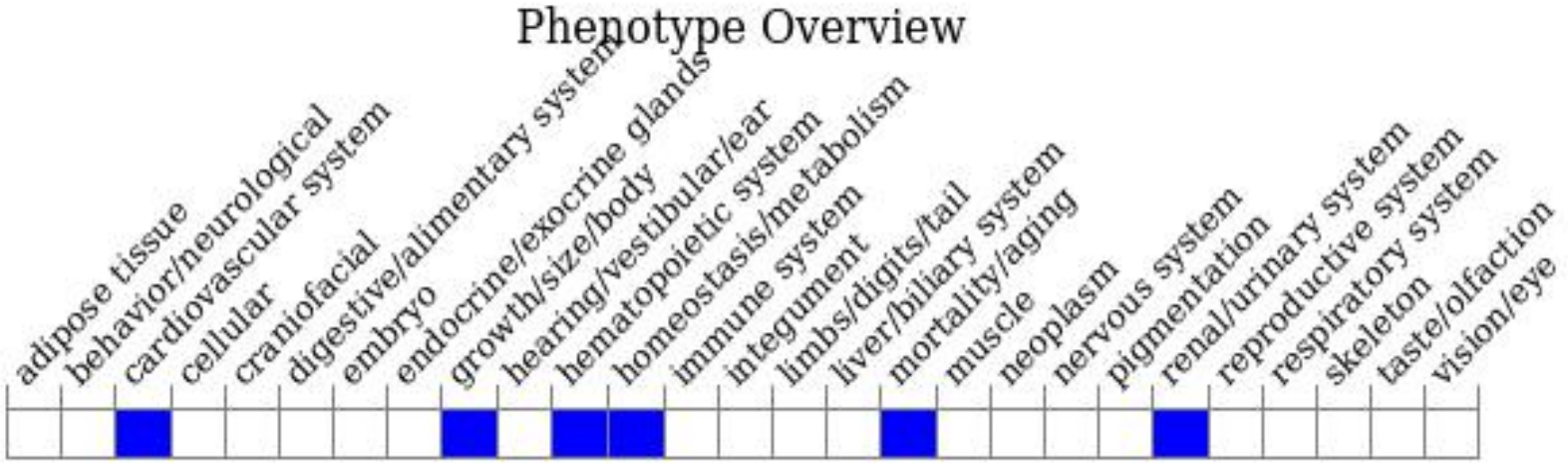
Genomic location distribution



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/).

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

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