Klk1 Cas9-KO Strategy

Designer: Yun Li

Reviewer: Shuang Zhang

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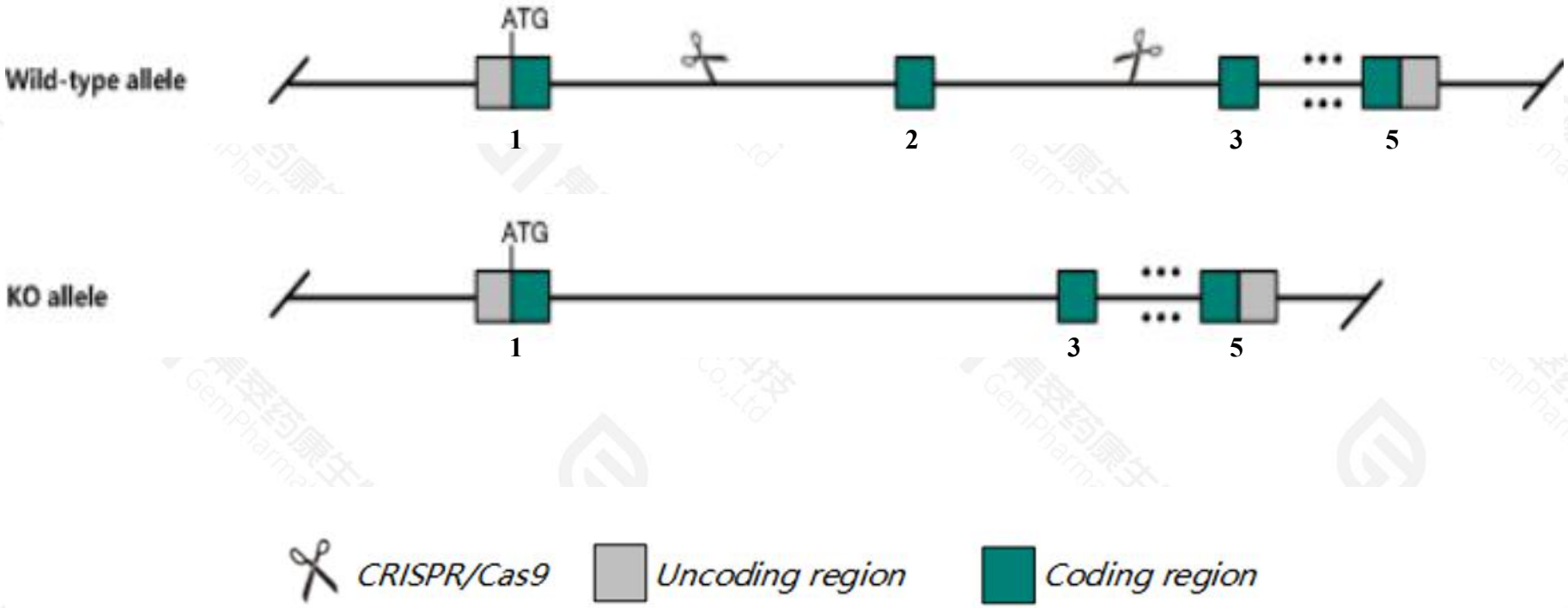
Project Overview

Project Name	***Klk1***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Klk1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Klk1* gene has 3 transcripts. According to the structure of *Klk1* gene, exon2 of *Klk1-201*(ENSMUST00000075162.5) transcript is recommended as the knockout region.The region contains 160bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Klk1* gene. The brief process is as follows: CRISPR/Cas9 system 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

➢ The *Klk1* gene is located on the Chr7. 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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Klk1 kallikrein 1 [Mus musculus (house mouse)]

Gene ID: 16612, updated on 25-Sep-2020

Summary

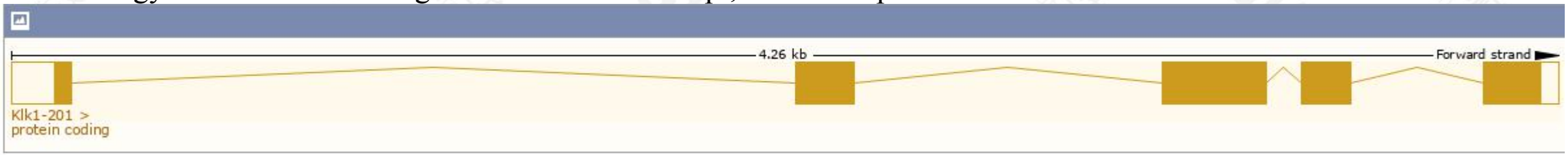
Official Symbol	Klk1 provided by MGI
Official Full Name	kallikrein 1 provided byMGI
Primary source	MGI:MGI:102850
See related	Ensembl:ENSMUSG00000063903
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	0610007D04Rik, KAL-B, Kal, Klk, Klk1b6, Klk6, mGk-6
Summary	This gene encodes a member of the kallikrein subfamily of serine proteases that are involved in diverse physiological functions such as skin desquamation, tooth enamel formation, seminal liquefaction, synaptic neural plasticity and brain function. The encoded preproprotein undergoes proteolytic cleavage of the activation peptide to generate the functional enzyme. Mice lacking the encoded protein are unable to generate significant levels of kinins in most tissues, develop cardiovascular abnormalities and exhibit hypercalciuria of renal origin. This gene is located in a cluster of several related kallikrein genes on chromosome 7. Alternative splicing results in multiple transcript variants encoding different isoforms, some of which may undergo similar processing. [provided by RefSeq, Feb 2016]
Expression	Biased expression in kidney adult (RPKM 1758.5), colon adult (RPKM 1588.3) and 4 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

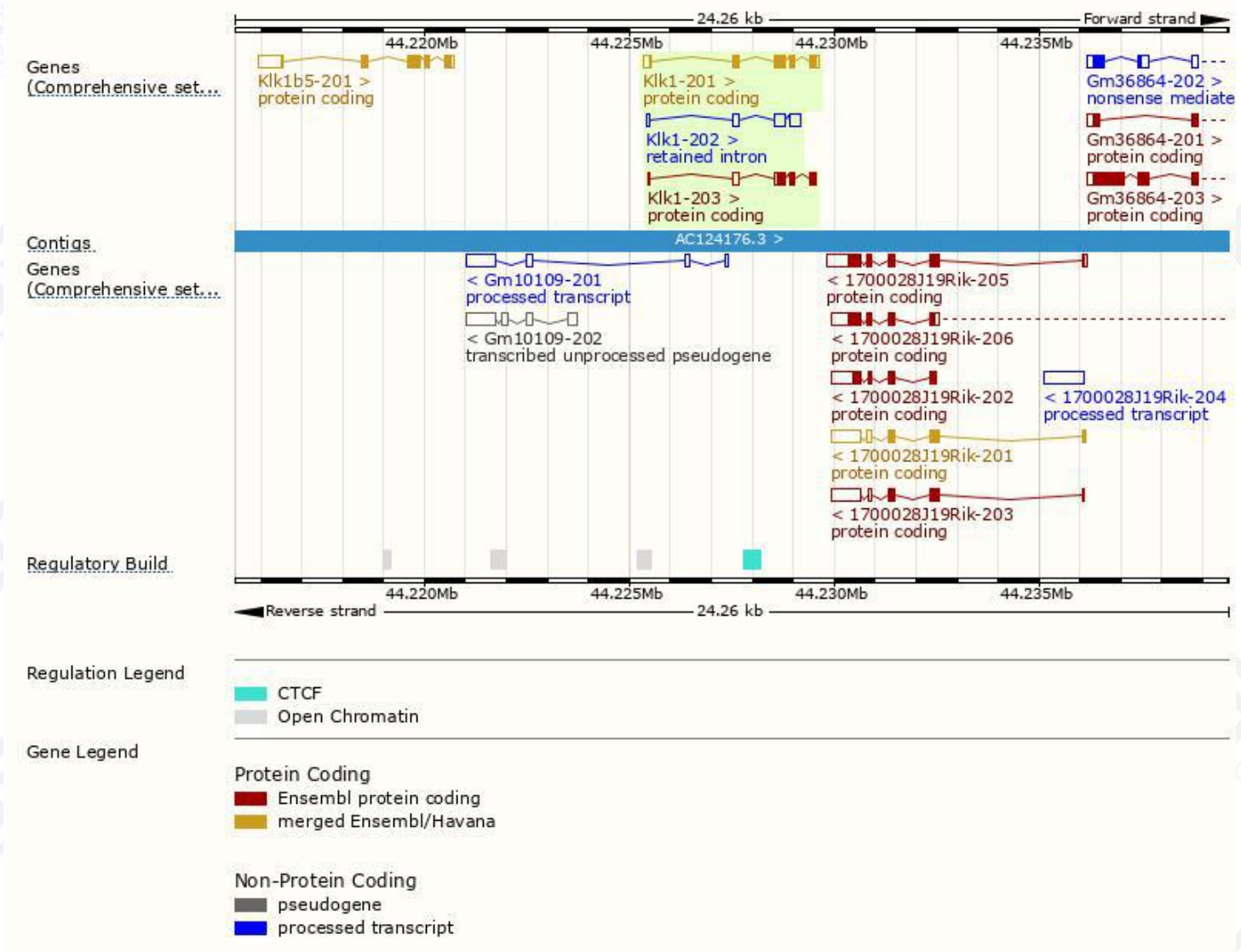
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Klk1-201	ENSMUST00000075162.5	954	261aa	Protein coding	CCDS21202		TSL:1 , GENCODE basic , APPRIS P1 ,
Klk1-203	ENSMUST00000206144.2	755	151aa	Protein coding	-		CDS 3' incomplete , TSL:3 ,
Klk1-202	ENSMUST00000205329.2	753	No protein	Retained intron	-		TSL:3 ,

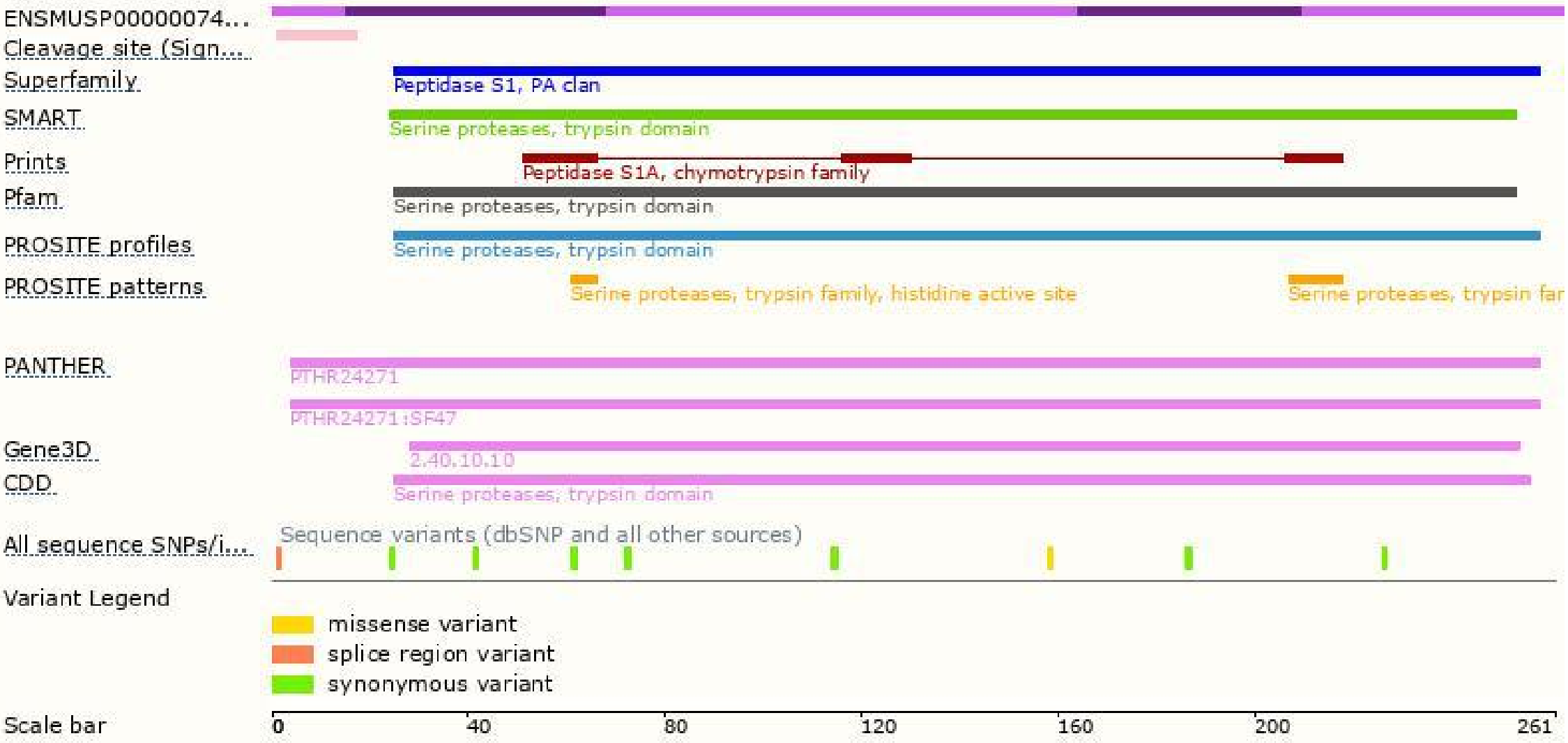
The strategy is based on the design of *Klk1-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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Tel: 400-9660890

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