Spink4 Cas9-KO *Strategy*

Designer: **Shilei Zhu**

Design Date: **2019-7-22**

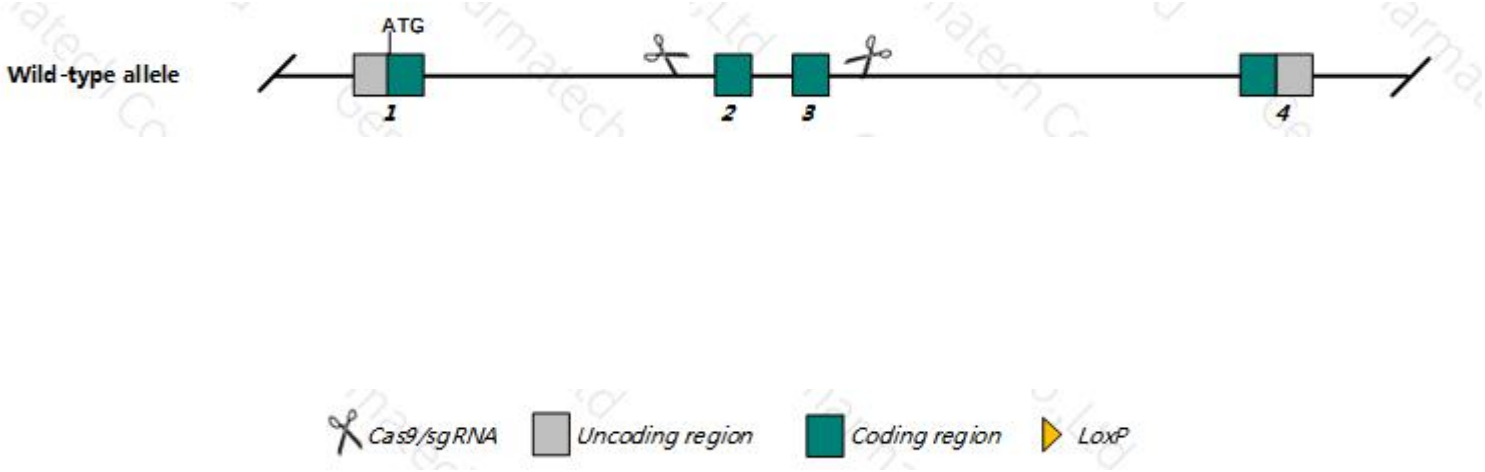
Project Overview

集萃药康 GemPharmatech

Project Name	***Spink4***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Spink4* gene. The schematic diagram is as follows:



Technical routes

集萃药康 GemPharmatech

- The *Spink4* gene has 2 transcript.According to the structure of *Spink4* gene, exon2-3 of *Spink4*-201 (ENSMUST00000030122.4) transcript is recommended as the knockout region.The region contains 154bp coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Spink4* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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.

Notice

- The *Spink4* gene is located on the Chr4. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information NCBI

集萃药康 GemPharmatech

Spink4 serine peptidase inhibitor, Kazal type 4 [*Mus musculus* (house mouse)]

Gene ID: 20731, updated on 31-Jan-2019

Summary

Official Symbol	Spink4 provided by MGI
Official Full Name	serine peptidase inhibitor, Kazal type 4 provided by MGI
Primary source	MGI:MGI:1341848
See related	Ensembl:ENSMUSG00000028415
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	MPGC60
Expression	Biased expression in large intestine adult (RPKM 3974.1), colon adult (RPKM 1771.4) and 3 other tissues See more
Orthologs	human all

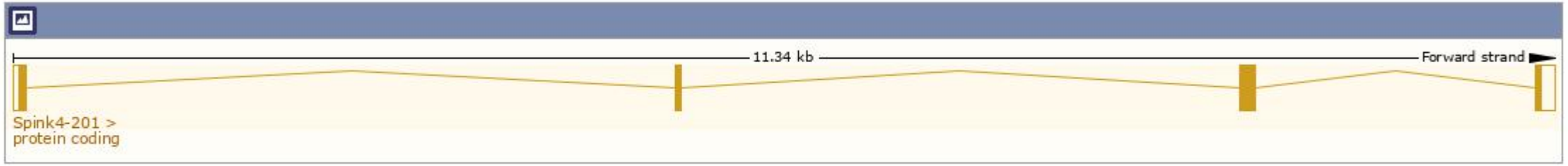
Transcript information Ensembl

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

Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Spink4-201	ENSMUST00000030122.4	397	86aa	Protein coding	CCDS18052	O35679	TSL:1 GENCODE basic APPRIS P1
Spink4-202	ENSMUST00000132943.1	223	No protein	lncRNA	-	-	TSL:5

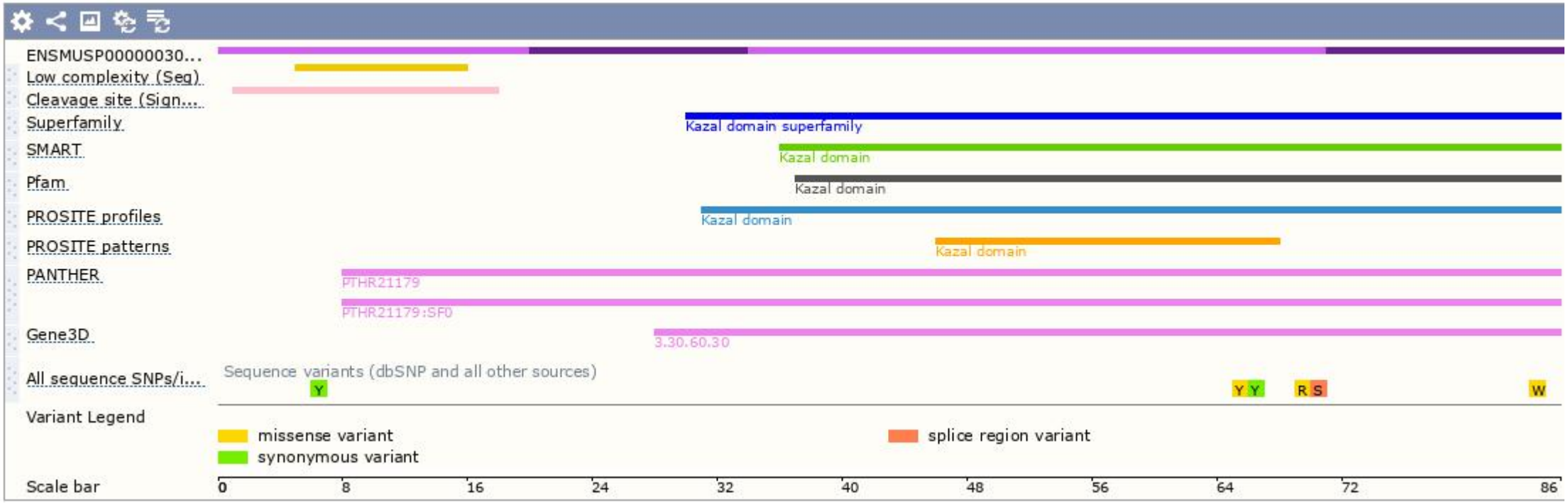
The strategy is based on the design of *Spink4*-201 transcript,The transcription is shown below



Statistics **Exons:** 4, **Coding exons:** 4, **Transcript length:** 397 bps, **Translation length:** 86 residues

Genomic location distribution

Protein domain



If you have any questions, you are welcome to inquire.
Tel: 400-9660890

集萃药康生物科技
GemPharmatech Co.,Ltd