

Col7a1 Cas9-KO Strategy

Designer: Huimin Su

Reviewer: Ruiuri Zhang

Design Date: 2020-6-28

Project Overview



Project Name

Col7a1

Project type

Cas9-KO

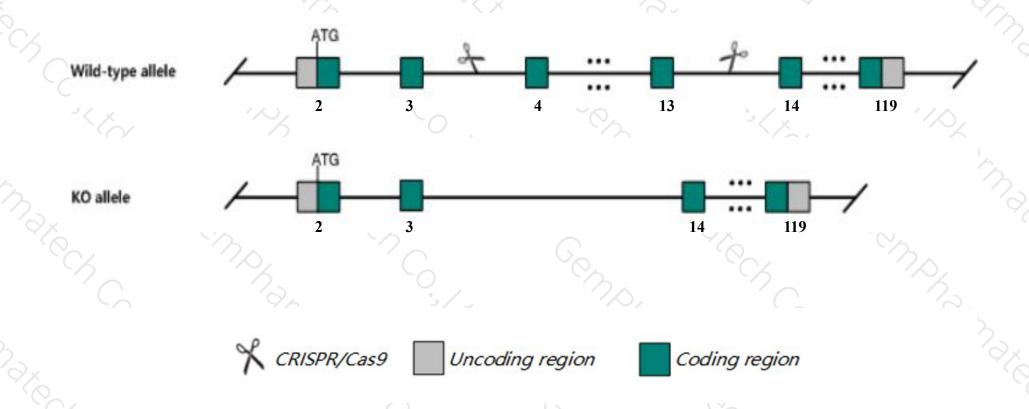
Strain background

C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Col7a1 gene. The schematic diagram is as follows:



Technical routes



- The *Col7a1* gene has 8 transcripts. According to the structure of *Col7a1* gene, exon4-exon13 of *Col7a1-202*(ENSMUST00000112070.7) transcript is recommended as the knockout region. The region contains 1370bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Col7a1* 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



- > According to the existing MGI data, mice homozygous for a knock-out allele are unable to reproduce and display postnatal growth retardation, blisters and erosion at sites of trauma, nonpigmented hair growth associated with hair loss, subepidermal blistering associated with poorly formed hemidesmosomes, and high postnatal lethality.
- > The *Col7a1* 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)



Col7a1 collagen, type VII, alpha 1 [Mus musculus (house mouse)]

Gene ID: 12836, updated on 26-Jun-2020

Summary

☆ ?

Official Symbol Col7a1 provided by MGI

Official Full Name collagen, type VII, alpha 1 provided by MGI

Primary source MGI:MGI:88462

See related Ensembl: ENSMUSG00000025650

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 AW209154

Expression Broad expression in limb E14.5 (RPKM 4.6), subcutaneous fat pad adult (RPKM 3.7) and 20 other tissues See more

Orthologs human all

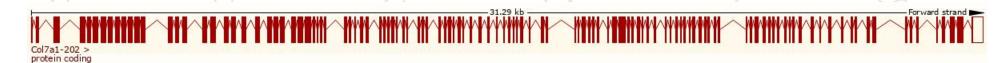
Transcript information (Ensembl)



The gene has 8 transcripts, all transcripts are shown below:

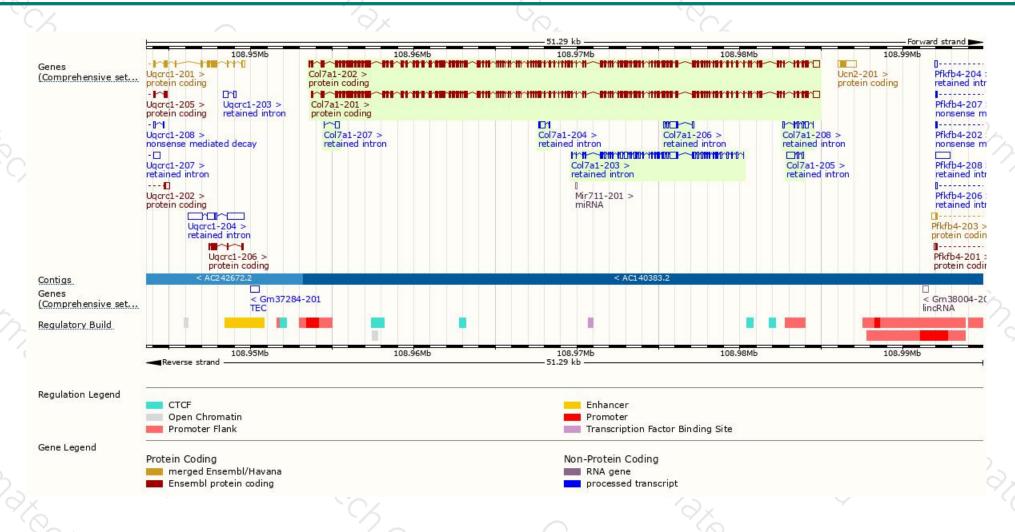
Name 🌲	Transcript ID	bp 🛊	Protein A	Biotype	CCDS	UniProt 🍦	Flags
Col7a1-202	ENSMUST00000112070.7	9250	2944aa	Protein coding	CCDS40770 ₽	Q63870 €	TSL:5 GENCODE basic APPRIS P1
Col7a1-201	ENSMUST00000026740.5	9198	<u>2944aa</u>	Protein coding	CCDS40770@	Q63870 €	TSL:1 GENCODE basic APPRIS P1
Col7a1-203	ENSMUST00000126780.7	2770	No protein	Retained intron	-	-	TSL:5
Col7a1-205	ENSMUST00000138588.1	695	No protein	Retained intron	120	28	TSL:5
Col7a1-206	ENSMUST00000149142.1	654	No protein	Retained intron	121	<u> </u>	TSL:3
Col7a1-208	ENSMUST00000198997.5	525	No protein	Retained intron		. 3	TSL:3
Col7a1-204	ENSMUST00000127905.1	341	No protein	Retained intron	1.00	<u> </u>	TSL:3
Col7a1-207	ENSMUST00000192424.1	245	No protein	Retained intron	5 5 8	-	TSL:5

The strategy is based on the design of *Col7a1-202* transcript, the transcription is shown below:



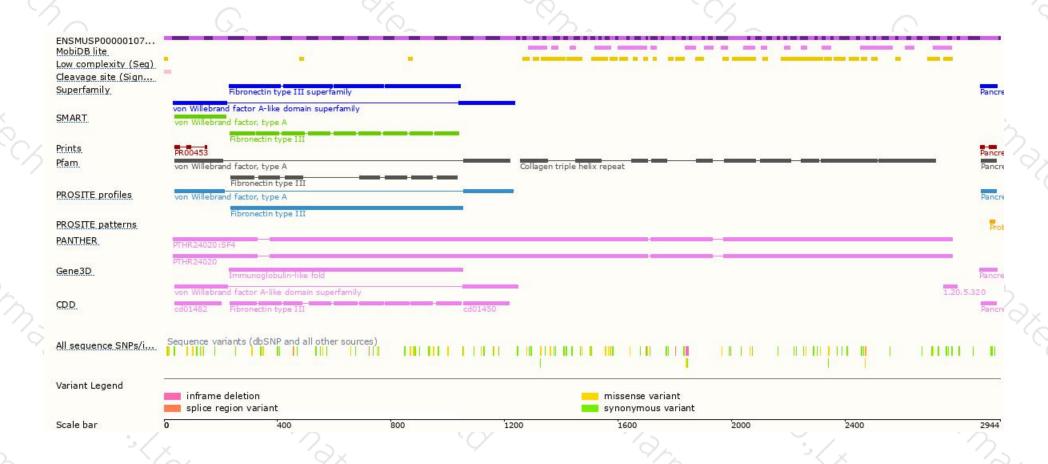
Genomic location distribution





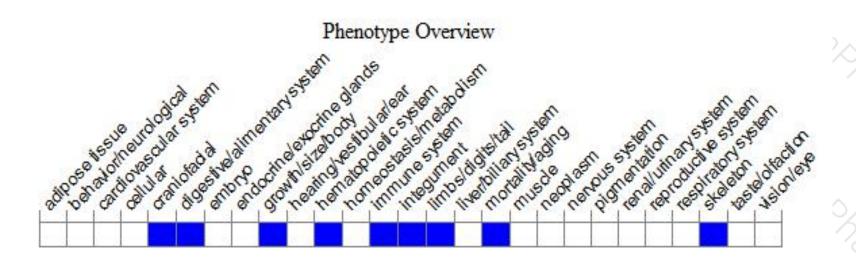
Protein domain





Mouse phenotype description(MGI)





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

According to the existing MGI data,mice homozygous for a knock-out allele are unable to reproduce and display postnatal growth retardation, blisters and erosion at sites of trauma, nonpigmented hair growth associated with hair loss, subepidermal blistering associated with poorly formed hemidesmosomes, and high postnatal lethality.



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





