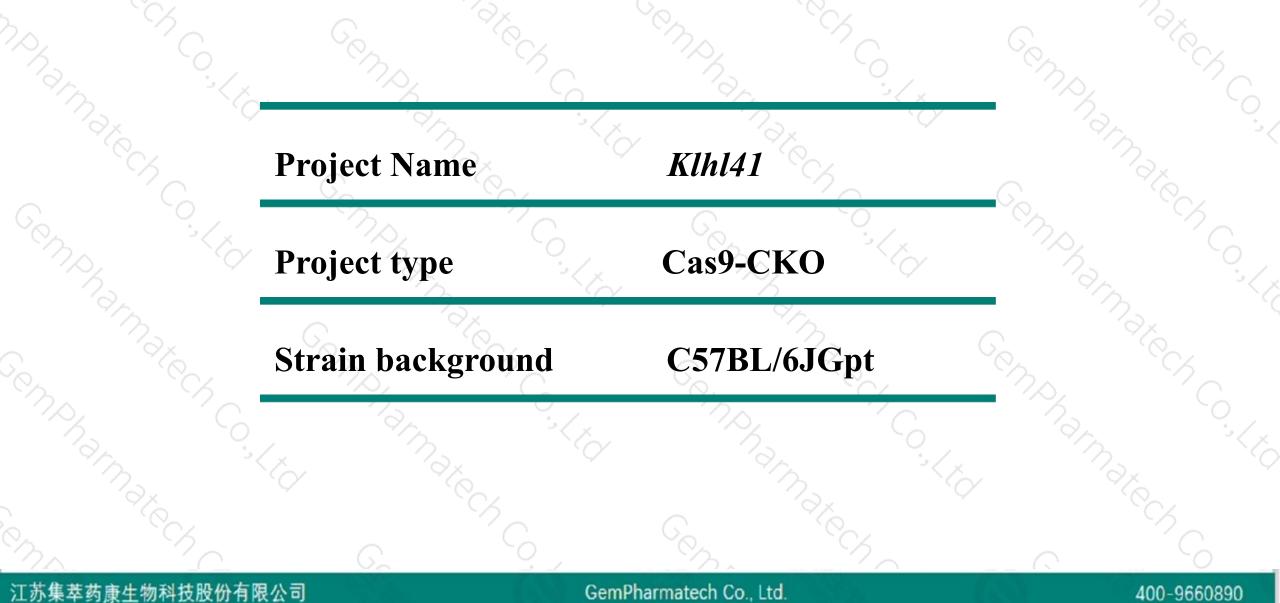


Klhl41 Cas9-CKO Strategy

Designer: Reviewer: Design Date: JiaYu Xiaojing Li 2020-3-5

Project Overview

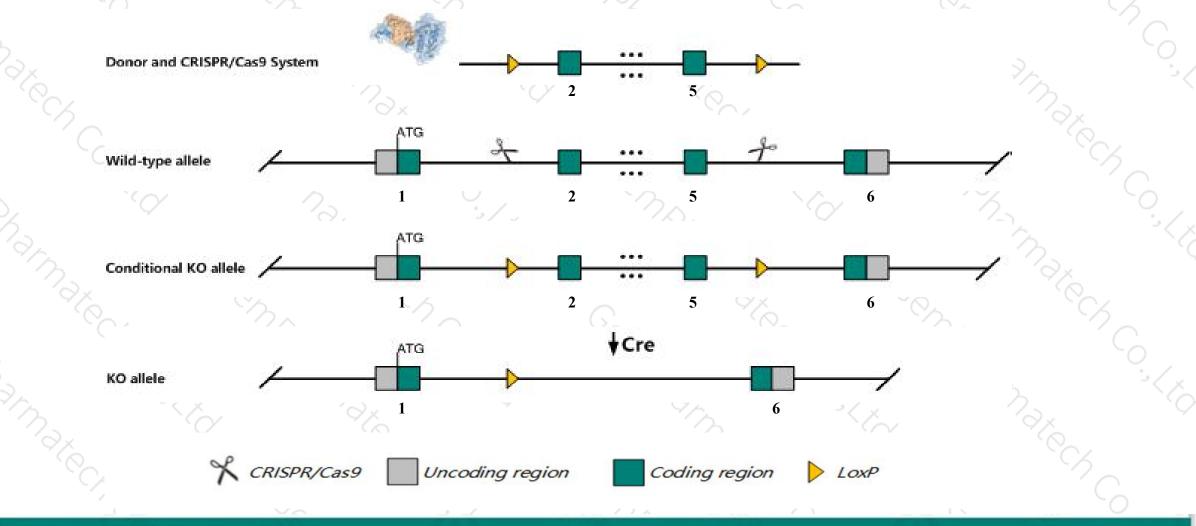




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Klhl41* gene. The schematic diagram is as follows:



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The Klhl41 gene has 1 transcript. According to the structure of Klhl41 gene, exon2-exon5 of Klhl41-201 (ENSMUST00000100050.3) transcript is recommended as the knockout region. The region contains 599bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Klhl41* gene. The brief process is as follows:CRISPR/Cas9 system 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.

The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice



According to the existing MGI data, The gene product is involved in stabilizing proteins involved in muscle function. Homozygous knockout affects the structure of muscle fibers and their sarcomeres, resulting in neonatal death.

> Some amino acids will remain at the N-terminus and some functions may be retained.

The Klhl41 gene is located on the Chr2. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

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Gene information (NCBI)



\$?

Klhl41 kelch-like 41 [Mus musculus (house mouse)]

Gene ID: 228003, updated on 31-Jan-2019

Summary

Official Symbol	KIhI41 provided by MGI
Official Full Name	kelch-like 41 provided byMGI
Primary source	MGI:MGI:2683854
See related	Ensembl:ENSMUSG0000075307
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	Gm112, Kbtbd10, SARCOSIN
Expression	Biased expression in heart adult (RPKM 9.8), mammary gland adult (RPKM 7.3) and 6 other tissues See more
Orthologs	human all

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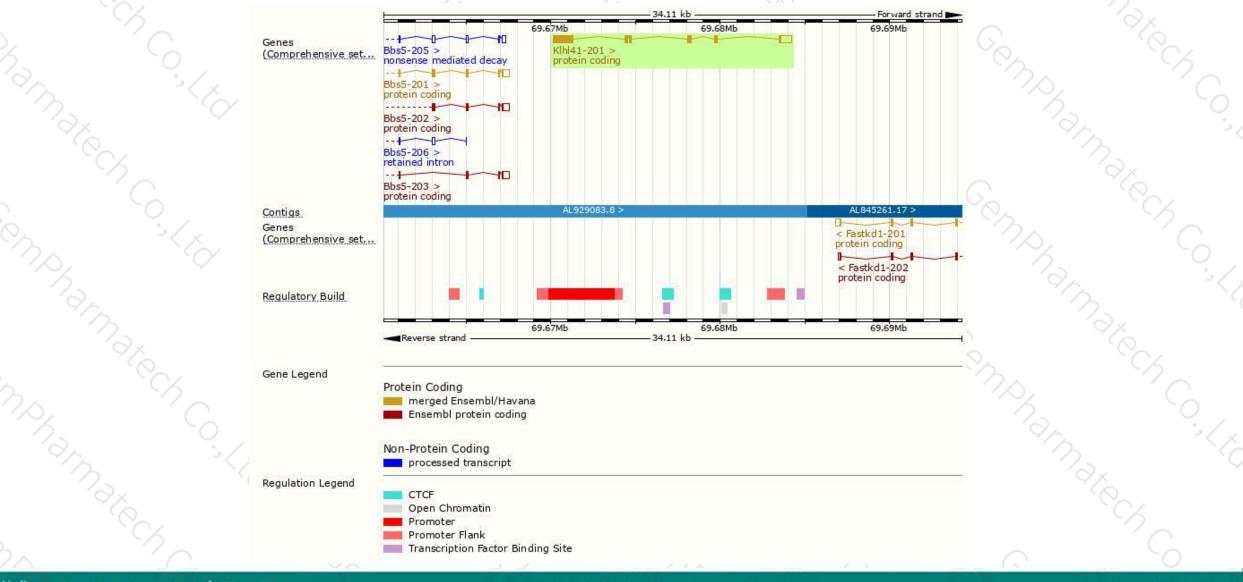
The gene has 1 transcript, and the transcript is shown below:

Name Transcript ID		cript ID bp Protein Biotype CCDS U				UniProt	Flags	5
Klhl41-201	ENSMUST00000100050.3 2529 600			Protein coding	CCDS38136	A2AUC9	TSL:1 GENCODE basic APPRIS P1	1.
Genphann Gennbhann Genn	SColler Comp	Sarn.					Cempton atende	
The strategy	is based on the design of	Klhl41	-201 tran	script,The trans	scription is sh	own below	Forward strand	
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Genomic location distribution



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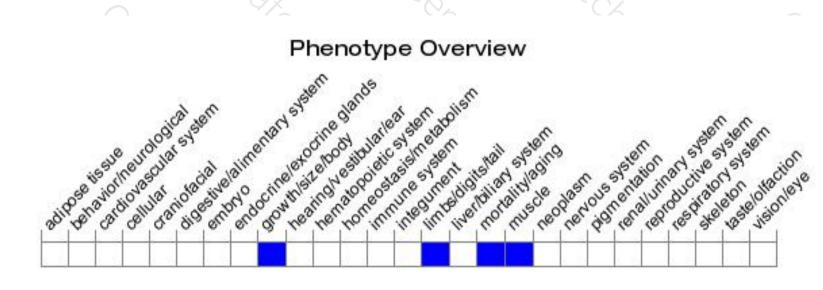
Protein domain



53.	ENSMUSP00000097 Low complexity (Seg) Superfamily	SKP1/BTB/POZ domain superfamily				Kelch-type beta propeller						-			
armar.	SMART	BTB/POZ dom	nain BTB/	Kelch-associat	ted			Kelch rep	peat type 1						
	<u>Pfam</u>	BTB/POZ domai	n BTB/	Kelch-associat	ed				Kelch repeat	type 1	-				
	PROSITE profiles PIRSF	BTB/POZ.dom BTB-kelch protein	nain									-	°ЧС		
	PANTHER	Kelch-like protein 41										_			
	Gene3D	PTHR24412 3.30.710.10	1.25,	40.420	-	Kelch	n-type b	eta prop	eller			_			
	CDD		cd	14735									S		
	All sequence SNPs/i	Sequence variants	s (dbSNP and	d all other so	urces)	n I	(T	Q,	I.	(\cdot)	1.1	4	<u>`</u> 0,		
1	Variant Legend	missense var											X		
	Scale bar	0 60	120	180	240	30	0	360	420	480	540	606			

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, The gene product is involved in stabilizing proteins involved in muscle function. Homozygous knockout affects the structure of muscle fibers and their sarcomeres, resulting in neonatal death.



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



