

# Klhl13 Cas9-KO Strategy

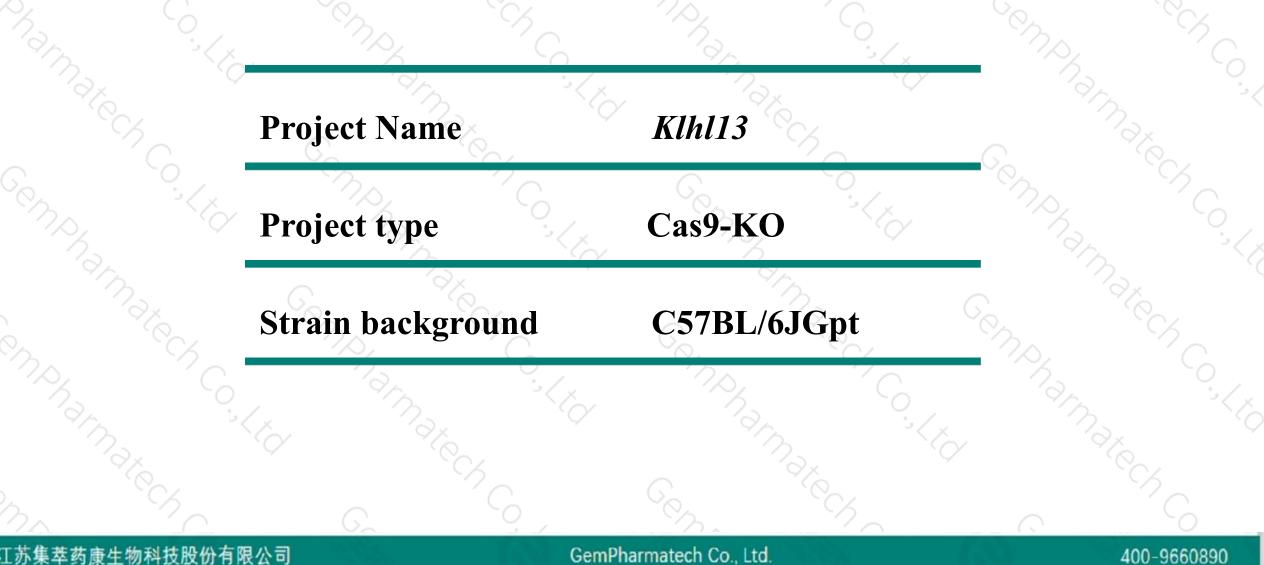
Designer: JiaYu

**Reviewer: Xiaojing Li** 

**Design Date: 2020-8-28** 

### **Project Overview**



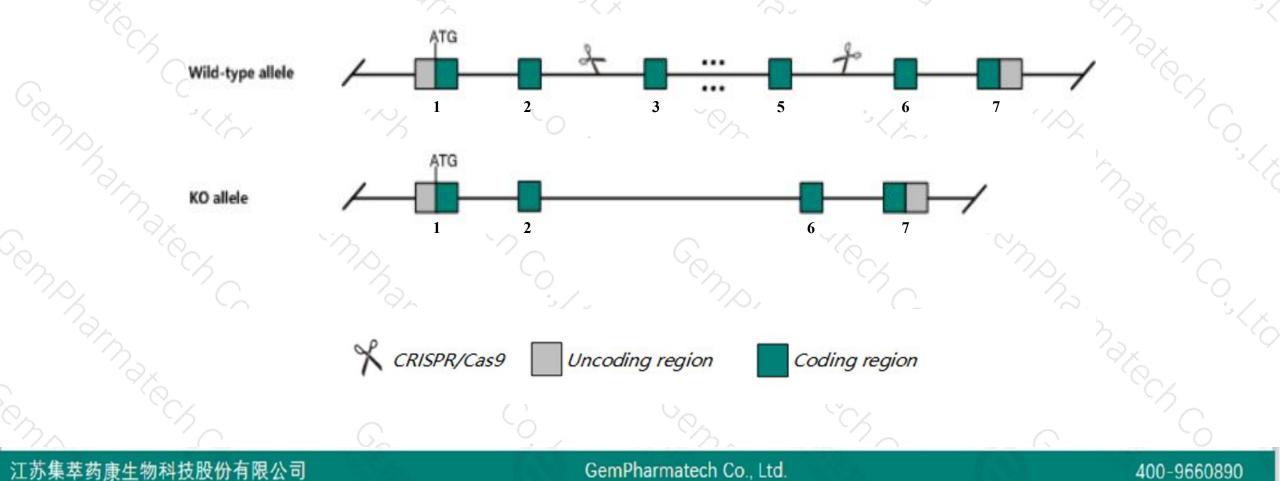


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### **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the *Klhl13* gene. The schematic diagram is as follows:





➤ The *Klhl13* gene has 7 transcripts. According to the structure of *Klhl13* gene, exon3-exon5 of *Klhl13-205*(ENSMUST00000115319.8) transcript is recommended as the knockout region. The region contains 1126bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Klhl13* 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.

- > The *Klhl13* gene is located on the ChrX. 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.

Notice

## **Gene information (NCBI)**



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#### Klhl13 kelch-like 13 [Mus musculus (house mouse)]

Gene ID: 67455, updated on 13-Mar-2020

#### Summary

Official Symbol	Kihi13 provided by MGI
<b>Official Full Name</b>	kelch-like 13 provided byMGI
<b>Primary source</b>	MGI:MGI:1914705
See related	Ensembl:ENSMUSG0000036782
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	1200009K10Rik, AI451128, Bklhd2, D130072F20Rik
Expression	Biased expression in placenta adult (RPKM 72.4), CNS E18 (RPKM 17.0) and 7 other tissuesSee more
Orthologs	human all

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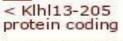
### **Transcript information (Ensembl)**



#### The gene has 7 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Klhl13-205	ENSMUST00000115319.8	3349	<u>648aa</u>	Protein coding	CCDS72348	<u>Q80TF4</u>	TSL:1 GENCODE basic APPRIS ALT1
Klhl13-203	ENSMUST00000115316.8	3064	<u>640aa</u>	Protein coding	CCDS81105	<u>Q80TF4</u>	TSL:5 GENCODE basic
Klhl13-201	ENSMUST0000035973.4	2996	<u>638aa</u>	Protein coding	CCDS40891	<u>Q80TF4</u>	TSL:5 GENCODE basic APPRIS P3
Klhl13-202	ENSMUST00000115313.7	3251	<u>604aa</u>	Protein coding	~	<u>Q80TF4</u>	TSL:1 GENCODE basic
Klhl13-204	ENSMUST00000115317.8	3216	<u>654aa</u>	Protein coding	-	<u>Q80TF4</u>	TSL:5 GENCODE basic APPRIS ALT1
Klhl13-206	ENSMUST00000144360.7	887	No protein	Processed transcript	5	-	TSL:2
Klhl13-207	ENSMUST00000153345.1	395	No protein	Processed transcript	-	-	TSL:3

The strategy is based on the design of *Klhl13-205* transcript, the transcription is shown below:



Reverse strand -

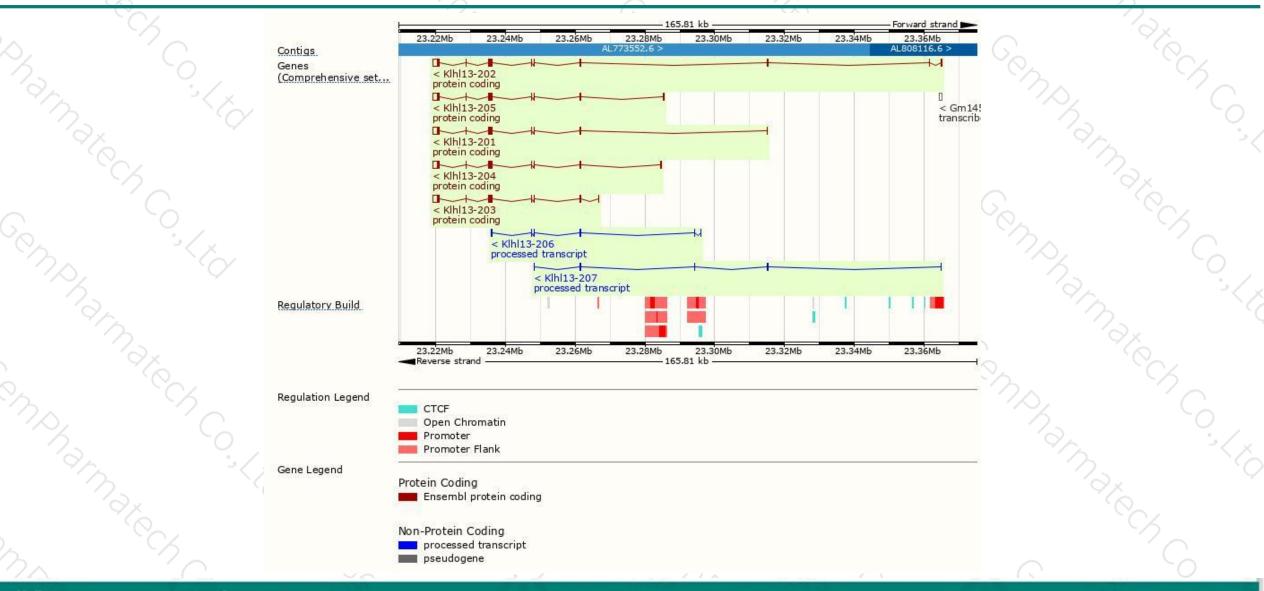
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66.28 kb

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### **Genomic location distribution**



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### **Protein domain**

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Superfamily		SKP1/B	STB/POZ dom	ain superfa	mily	Ke	elch-type be	eta propeller			2
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Gene 3D.     3.30.710.10       All sequence SNPs/i     Sequence variants (dbSNP and all others)	Superfamily.     SKP1/BTB/POZ domain superfamily       SMART.     BTB/POZ domain       Pfam.     BTB/POZ domain       Pfam.     BTB/POZ domain       PROSITE profiles.     BTB/POZ domain       PIRSE.     BTB/POZ domain       PANTHER.     PTHR45632       PTHR45632 (SF7       Gene3D.     3.30.710.10       CDD.     cd18239       All sequence SNPs/i     Sequence variants (dbSNP and all other sources)	Superfamily     SKP1/BTB/POZ domain superfamily     Ke       SMART.     BTB/POZ domain     BTB/Kelch-associated     Ke       Pfam.     BTB/POZ domain     BTB/Kelch-associated     Ke       PROSITE profiles     BTB/POZ domain     BTB/Kelch-associated     Ke       PROSITE profiles     BTB-kelch protein     PROSITE profiles     Kelch       PANTHER.     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BTB/PO2 domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/PO2 domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/PO2 domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/PO2 domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     PTHR45632 (SF7     Gene3D     Gene3D     Kelch repeat type 1       COD     cd18449     cd18449     Gene3D     Gene3D	Superfamily     SKP1/BTB/POZ domain superfamily     Kelch-type beta propeller       SMART.     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BTB/POZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/POZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/POZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/Kelch-associated     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/ROZ domain     Kelch repeat type 1       PROSITE profiles     BTB/ROZ domain     BTB/ROZ domain     F       PIRSF     BTB-kelch protein     F     F       Gene3D.     3.30.710.10     1.25.40.420     Kelch-type beta propeller       CDD.     cd18239     Cd18239     Cd18239



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



