

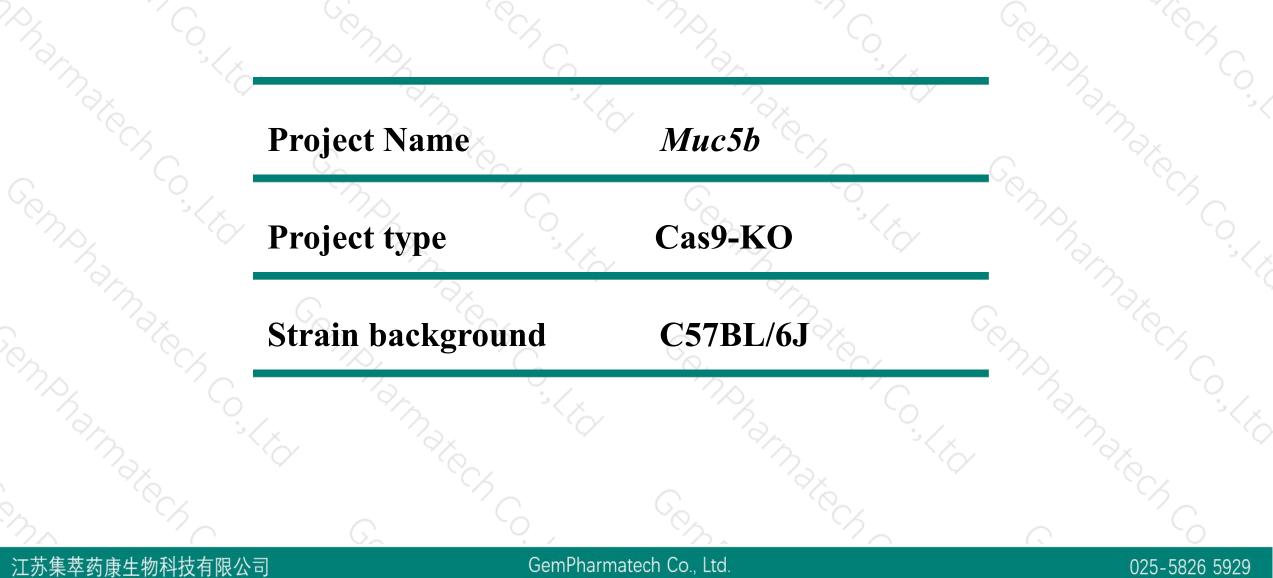
# Muc5b Cas9-KO Strategy

Cemphamatech ( Companyated Co. Designer:Qiong Zhou

enphamatech,

## **Project Overview**





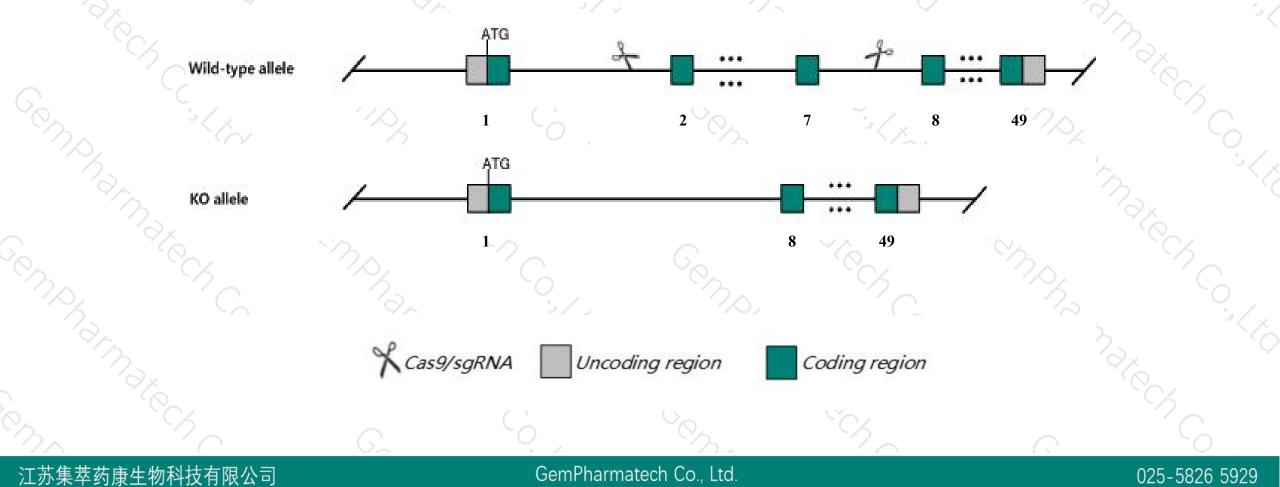
江苏集萃药康生物科技有限公司

GemPharmatech Co., Ltd.

# **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the Muc5b gene. The schematic diagram is as follows:





- The Muc5b gene has 1 transcript. According to the structure of Muc5b gene, exon2-exon7 of Muc5b-201 ( ENSMUST00000165147.2) transcript is recommended as the knockout region. The region contains 716bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Muc5b* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

025-5826 5929

- According to the existing MGI data, Mice homozygous for a knock-out allele accumulate materials in the upper and lower airways leading to chronic infection and inflammation that does not resolve and results in premature death. Macrophage function is impaired.
- The Muc5b 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Notice

# **Gene information (NCBI)**



\$ ?

## Muc5b mucin 5, subtype B, tracheobronchial [Mus musculus (house mouse)]

Gene ID: 74180, updated on 31-Jan-2019

### Summary

Official Symbol	Muc5b provided by MGI
Official Full Name	mucin 5, subtype B, tracheobronchial provided byMGI
Primary source	MGI:MGI:1921430
See related	Ensembl:ENSMUSG0000066108
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	2300002I04Rik, A130042M24, AV085033, MUC5, MUC9, mucin 5b
Expression	Biased expression in genital fat pad adult (RPKM 37.9) and lung adult (RPKM 11.8)See more
Orthologs	human all

#### 江苏集萃药康生物科技有限公司

GemPharmatech Co., Ltd.



# **Transcript information (Ensembl)**



Forward strand

025-5826 5929

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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Muc5b-201	ENSMUST00000165147.2	14964	<u>4800aa</u>	Protein coding	CCDS52448	E9Q513	TSL:5 GENCODE basic APPRIS P1
°°	4	$\sim$	)x	$\sim$	6	,	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

The strategy is based on the design of Muc5b-201 transcript, The transcription is shown below

Muc5b-201 > protein coding

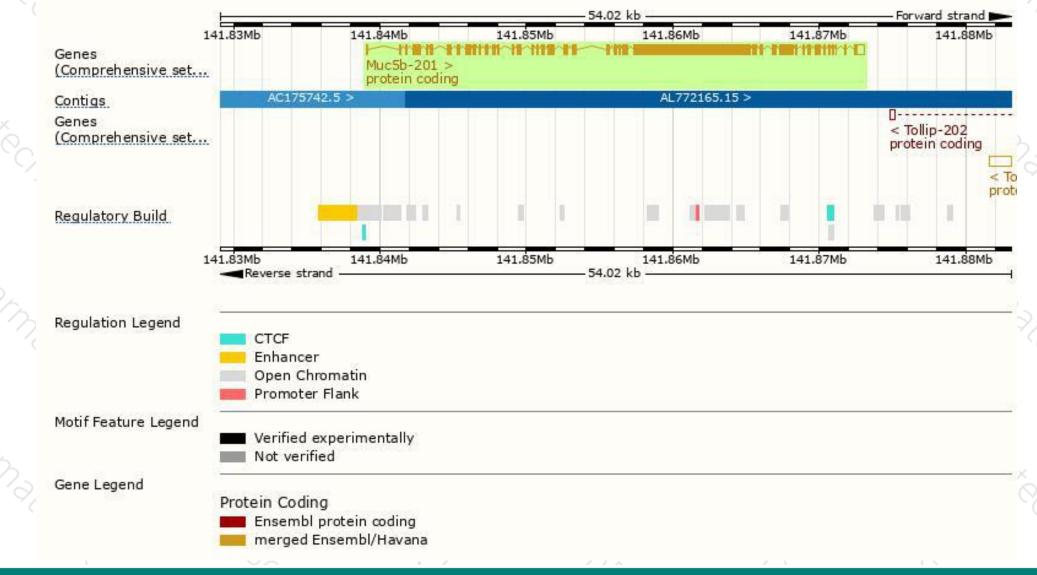
## 江苏集萃药康生物科技有限公司

GemPharmatech Co., Ltd.

34.02 kb

## **Genomic location distribution**





#### 江苏集萃药康生物科技有限公司

GemPharmatech Co., Ltd.

## 025-5826 5929

## **Protein domain**



ENSMUSP00000128... MobiDB lite Low complexity (Seg) Cleavage site (Sign... hmmpanther PTHR11339:SF310 FTHR 11339 Serine protease inhibitor-like superfamily Superfamily domains SSF57603 SMART domains Uncharacterised domain, cysteine-rich von Willebrand factor, type D domain WVFC domain VWFC domain Pfam domain WxxW domain Trypsin Inhibitor-like, cysteine rich domain von Willebrand factor, type D domain Uncharacterised domain, cysteine-rich PROSITE profiles von Willebrand factor, type D domain VWFC dorr PROSITE patterns WFC don Gene3D 2.10.25.10 Sequence variants (dbSNP and all other sources All sequence SNPs/i... Variant Legend inframe insertion missense variant splice region variant synonymous variant Scale bar 600 1200 1800 2400 3000 3600 4200 4800

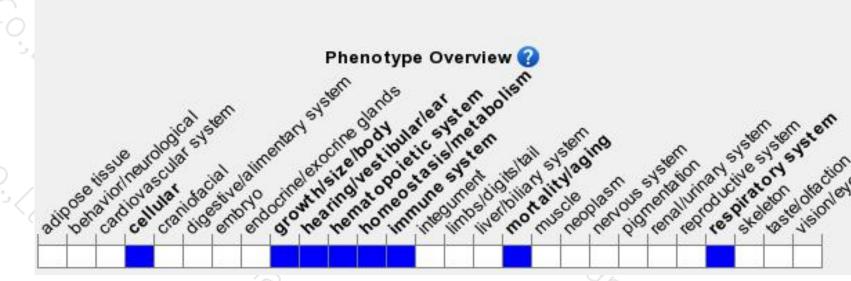
江苏集萃药康生物科技有限公司

GemPharmatech Co., Ltd.



## 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 accumulate materials in the upper and lower airways leading to chronic infection and inflammation that does not resolve and results in premature death. Macrophage function is impaired.

## 江苏集萃药康生物科技有限公司

## GemPharmatech Co., Ltd

#### 025-5826 5929



If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



