

# Scmh1 Cas9-CKO Strategy

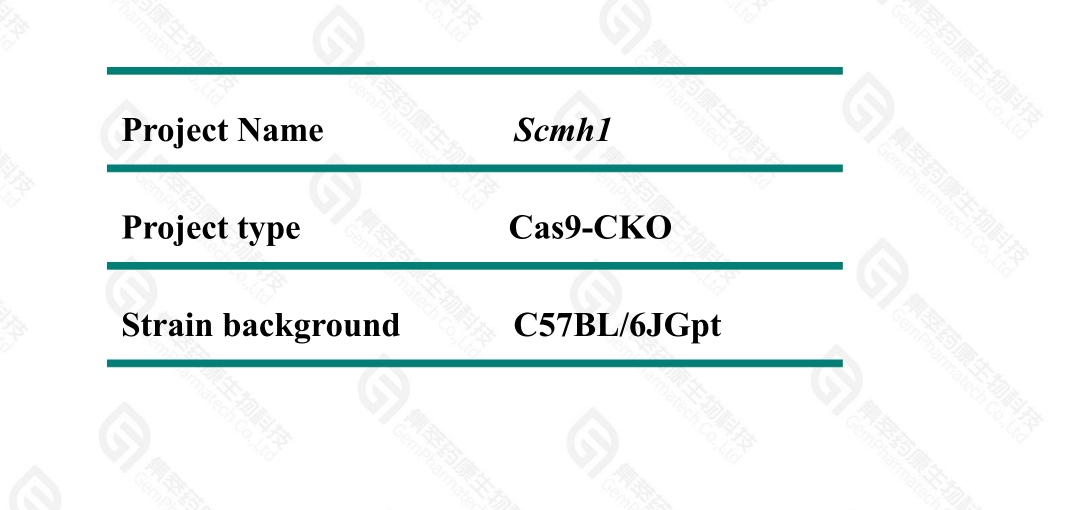
**Designer: Shanhong Tao** 

**Reviewer: Longyun Hu** 

**Design Date: 2022-5-27** 

# **Project Overview**



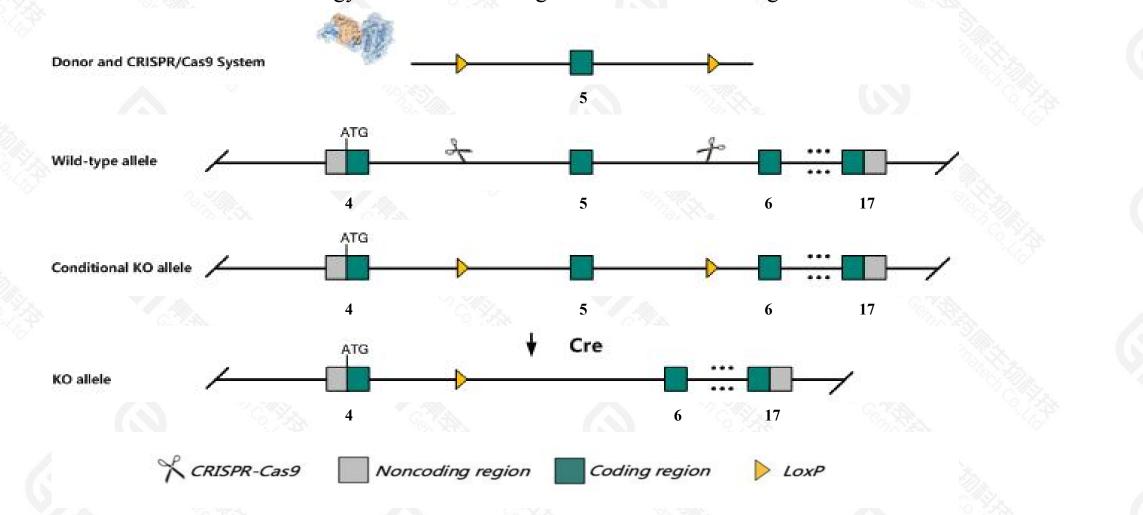


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# **Conditional Knockout strategy**

This model will use CRISPR-Cas9 technology to edit the *Scmh1* gene. The schematic diagram is as follows:



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### **Technical routes**



 The Scmh1 gene has 14 transcripts. According to the structure of Scmh1 gene, exon5 of Scmh1-204(ENSMUST00000106301.8) transcript is recommended as the knockout region. The region contains 71bp coding sequence.
 Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR-Cas9 technology to modify *Scmh1* 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.



- According to the existing MGI data, mice homozygous for an allele lacking the SPM domain exhibit partial penetrance of posterior vertebral transformations and male infertility with azoospermia and arrest of spermatogenesis. Mice homozygous for a knock-out allele exhibit abnormal hematopoiesis but normal fertility and skeleton.
  Transcript *Scmh1-205* may not be affected.
- > The *Scmh1* 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

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# Gene information (NCBI)

### Scmh1 sex comb on midleg homolog 1 [Mus musculus (house mouse)]

Gene ID: 29871, updated on 28-Apr-2022

### - Summary

Official Symbol	Scmh1 provided by MGI
<b>Official Full Name</b>	sex comb on midleg homolog 1 provided by <u>MGI</u>
Primary source	MGI:MGI:1352762
See related	Ensembl:ENSMUSG000000085
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	AI315320, AI851618, Scml1, Scml3
Expression	Ubiquitous expression in testis adult (RPKM 9.0), bladder adult (RPKM 6.2) and 28 other tissues See more
Orthologs	human all



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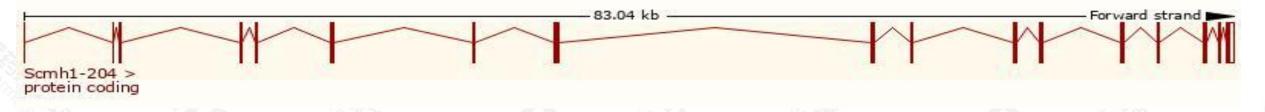
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# **Transcript information (Ensembl)**

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Scmh1-201	ENSMUST0000000087.13	3353	<u>664aa</u>	Protein coding	CCDS18589		TSL:1 , GENCODE basic , APPRIS P3 ,
Scmh1-204	ENSMUST00000106301.8	2584	<u>706aa</u>	Protein coding	CCDS51292		TSL:1 , GENCODE basic , APPRIS ALT2
Scmh1-202	ENSMUST0000064991.13	2548	<u>706aa</u>	Protein coding	CCDS51292		TSL:5 , GENCODE basic , APPRIS ALT2
Scmh1-203	ENSMUST00000106298.10	1995	<u>664aa</u>	Protein coding	CCDS18589		TSL:1, GENCODE basic, APPRIS P3,
Scmh1-206	ENSMUST00000132116.8	623	<u>28aa</u>	Protein coding	¥.		CDS 3' incomplete , TSL:5 ,
Scmh1-205	ENSMUST00000122860.2	530	<u>177aa</u>	Protein coding			CDS 5' and 3' incomplete , TSL:3 ,
Scmh1-211	ENSMUST00000144555.8	3544	No protein	Processed transcript	-		TSL:1,
Scmh1-214	ENSMUST00000153099.2	1776	No protein	Processed transcript	-		TSL:1,
Scmh1-210	ENSMUST00000136801.8	1536	No protein	Processed transcript	-		TSL:1,
Scmh1-209	ENSMUST00000134375.8	1132	No protein	Processed transcript	-		TSL:1,
Scmh1-212	ENSMUST00000144862.2	595	No protein	Processed transcript	2		TSL:3,
Scmh1-208	ENSMUST00000133290.2	557	No protein	Processed transcript	-		TSL:2,
Scmh1-207	ENSMUST00000133169.2	443	No protein	Processed transcript	Ξ.		TSL:2,
Scmh1-213	ENSMUST00000151345.8	352	No protein	Processed transcript			TSL:3 ,
	10 <sup>-2</sup>		1000	N. 2.			SV2.3

### The gene has 14 transcripts, all transcripts are shown below:

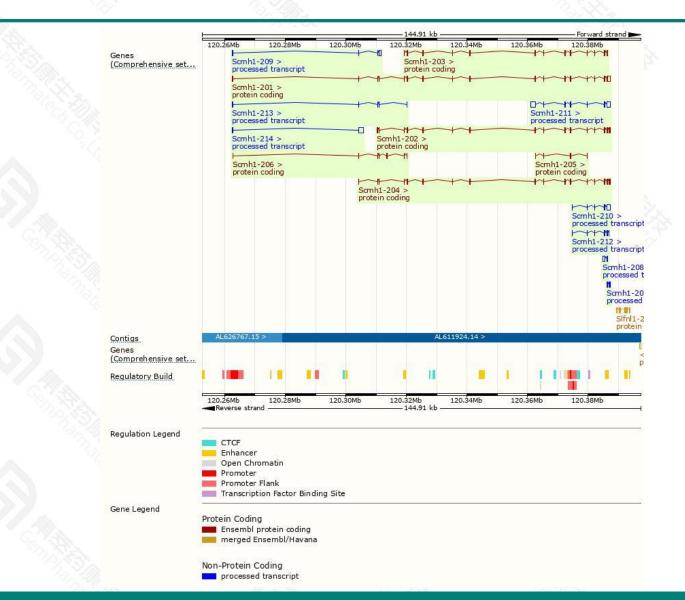
The strategy is based on the design of *Scmh1-204* transcript, the transcription is shown below:



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### **Genomic location distribution**



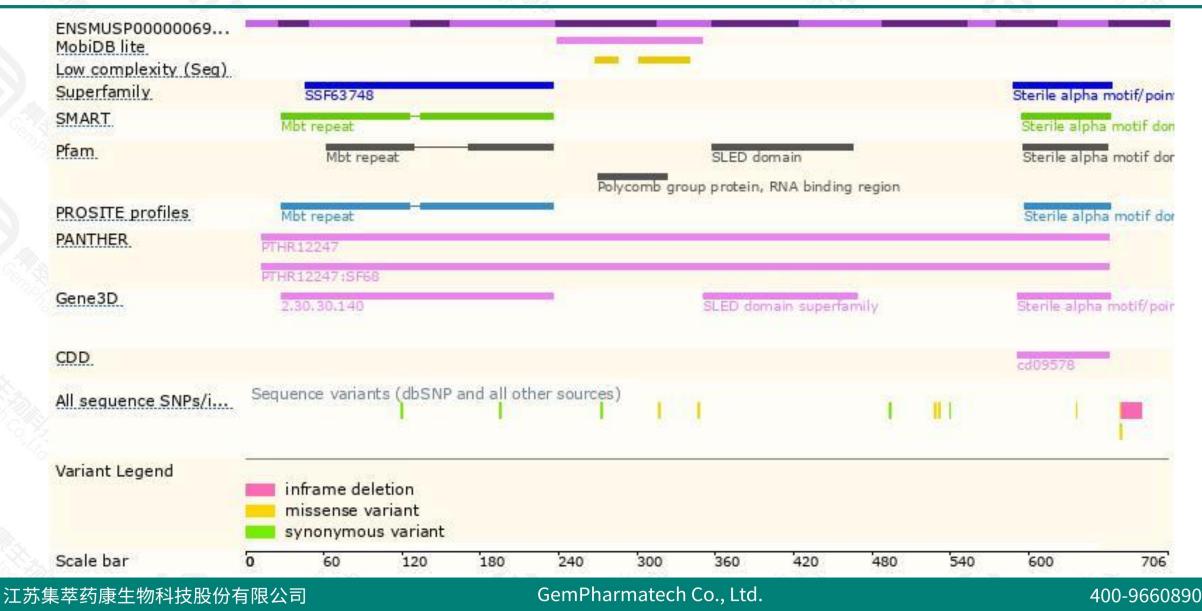


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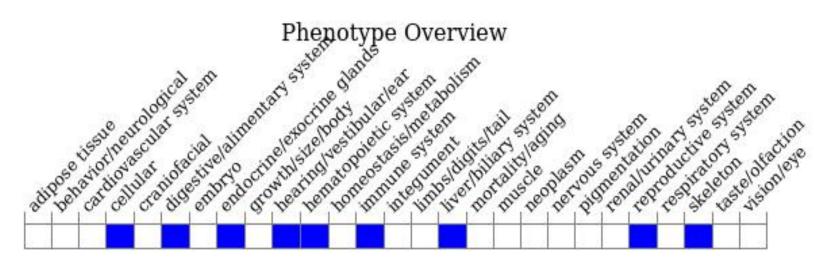
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### **Protein domain**





### 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 an allele lacking the SPM domain exhibit partial penetrance of posterior vertebral transformations and male infertility with azoospermia and arrest of spermatogenesis. Mice homozygous for a knock-out allele exhibit abnormal hematopoiesis but normal fertility and skeleton.

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



