

Usp19 Cas9-CKO Strategy

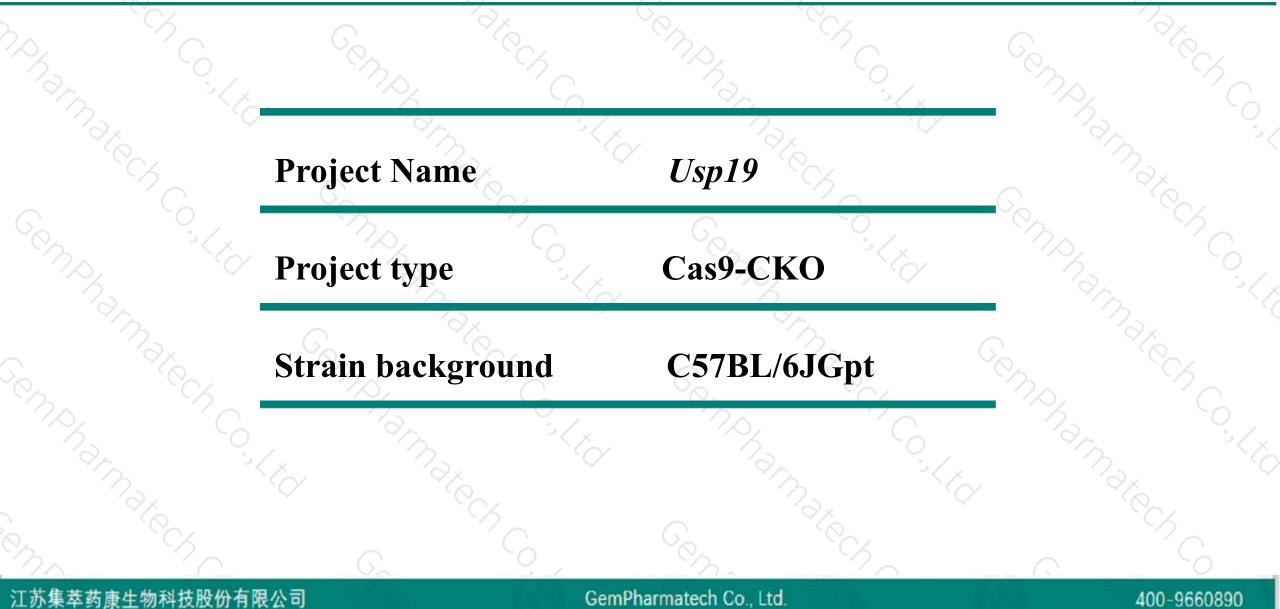
Designer: Reviewer:

Design Date:

Longyun Hu Jiayuan Yao 2019-11-29

Project Overview

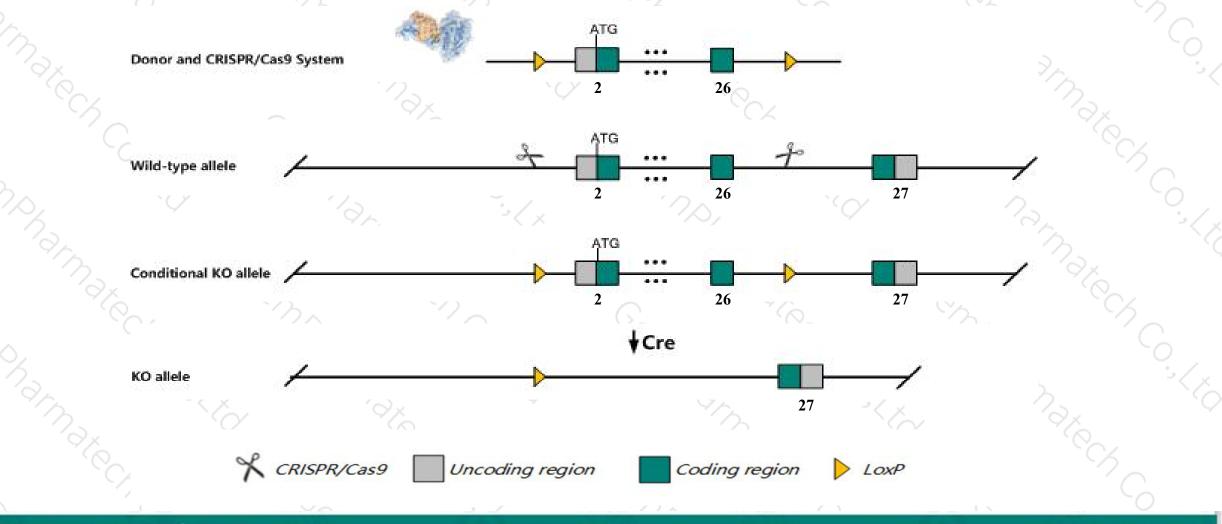




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Usp19 gene. The schematic diagram is as follows:



江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.



The Usp19 gene has 16 transcripts. According to the structure of Usp19 gene, exon2-exon26 of Usp19-201 (ENSMUST0000006854.12) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify Usp19 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 a null allele exhibit decreased body weight, reduced male fertility, and increased resistance to skeletal muscle atrophy induced by both glucocorticoids and denervation.
- The Usp19 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)



< ?

Usp19 ubiquitin specific peptidase 19 [Mus musculus (house mouse)]

Gene ID: 71472, updated on 2-Mar-2019

Summary

Official SymbolUsp19 provided by MGIOfficial Full NameMci.Mci.1918722Primary sourceMci.Mci.1918722See relatedEnsembl:ENSMUSG0000006676Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Rodentia; Myomorpha;
Muroidea; Murinae; Mus; MusAlso knowna8430421107Rik, Al047774, Zmynd9ExpressionUbiquitous expression in adrenal adult (RPKM 57.6), ovary adult (RPKM 50.1) and 28 other tissues
See more

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

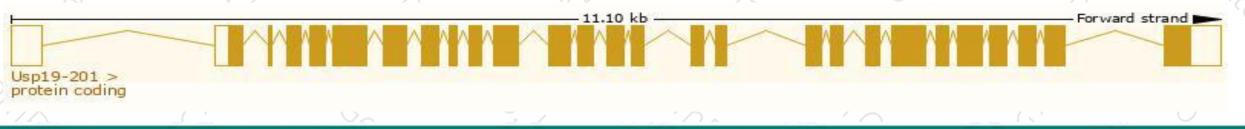
Transcript information (Ensembl)



The gene has 16 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	1
Usp19-201	ENSMUST0000006854.12	4791	<u>1360aa</u>	Protein coding	CCDS23529	Q3UJD6	TSL:1 GENCODE basic APPRIS P3	
Usp19-202	ENSMUST0000085044.13	4520	<u>1323aa</u>	Protein coding	CCDS52924	Q3UJD6	TSL:1 GENCODE basic APPRIS ALT2	
Usp19-210	ENSMUST00000193678.5	4517	<u>1322aa</u>	Protein coding	CCDS81075	A0A0A6YWX1	TSL:1 GENCODE basic APPRIS ALT2	
Usp19-204	ENSMUST00000178075.7	4512	<u>1324aa</u>	Protein coding	CCDS57702	J3KMM1	TSL:1 GENCODE basic APPRIS ALT2	
Usp19-203	ENSMUST00000166103.8	4473	<u>1299aa</u>	Protein coding	CCDS52925	E9Q9M5	TSL:1 GENCODE basic APPRIS ALT2	
Usp19-212	ENSMUST00000194171.5	3252	<u>991aa</u>	Protein coding	-	A0A0A6YWN9	CDS 5' incomplete TSL:1	
Usp19-215	ENSMUST00000194863.1	885	<u>295aa</u>	Protein coding	-	A0A0A6YX42	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3	
Usp19-208	ENSMUST00000193558.1	384	<u>128aa</u>	Protein coding	2	A0A0A6YXT9	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:1	D
Usp19-211	ENSMUST00000193975.1	1548	No protein	Retained intron	-	(27)	TSL:NA	1
Usp19-205	ENSMUST00000192854.5	1352	No protein	Retained intron	-	(. .)	TSL:5	
Usp19-214	ENSMUST00000194499.1	803	No protein	Retained intron	-	(12)	TSL:1	
Usp19-207	ENSMUST00000193412.1	753	No protein	Retained intron	-	828	TSL:3	
Usp19-216	ENSMUST00000195763.1	744	No protein	Retained intron	-	(27)	TSL:3	
Usp19-209	ENSMUST00000193571.1	729	No protein	Retained intron	-	1.0	TSL:2	
Usp19-213	ENSMUST00000194225.1	607	No protein	Retained intron	-	(2)	TSL:5	
Usp19-206	ENSMUST00000193183.5	525	No protein	Retained intron	-	1021	TSL:2	
		/		1				3

The strategy is based on the design of Usp19-201 transcript, The transcription is shown below

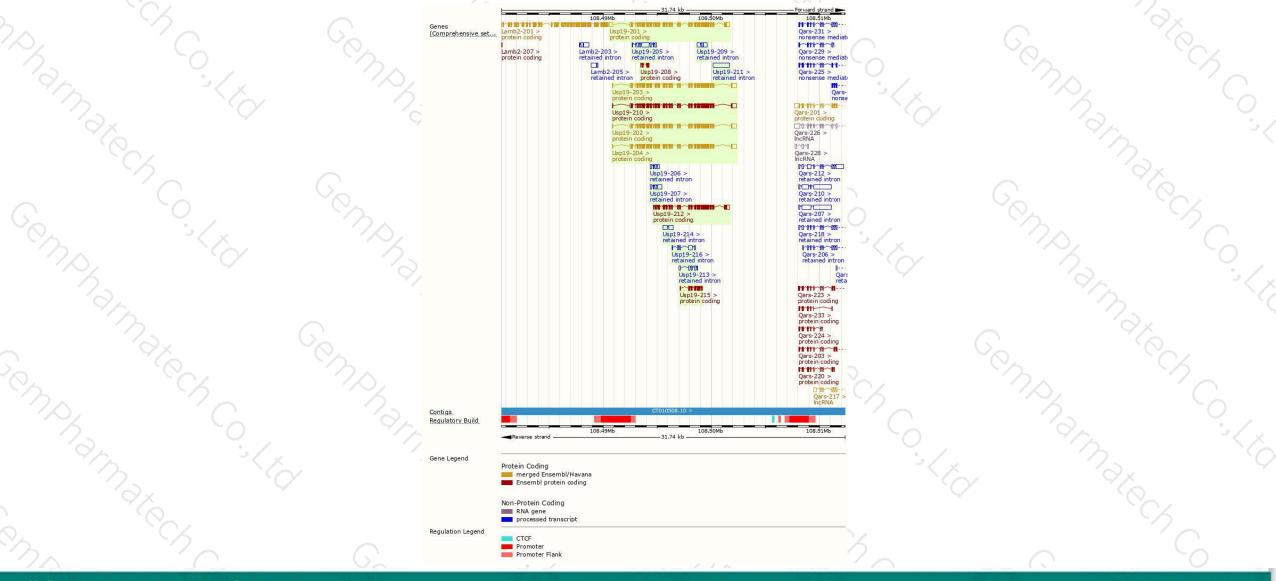


江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

Genomic location distribution



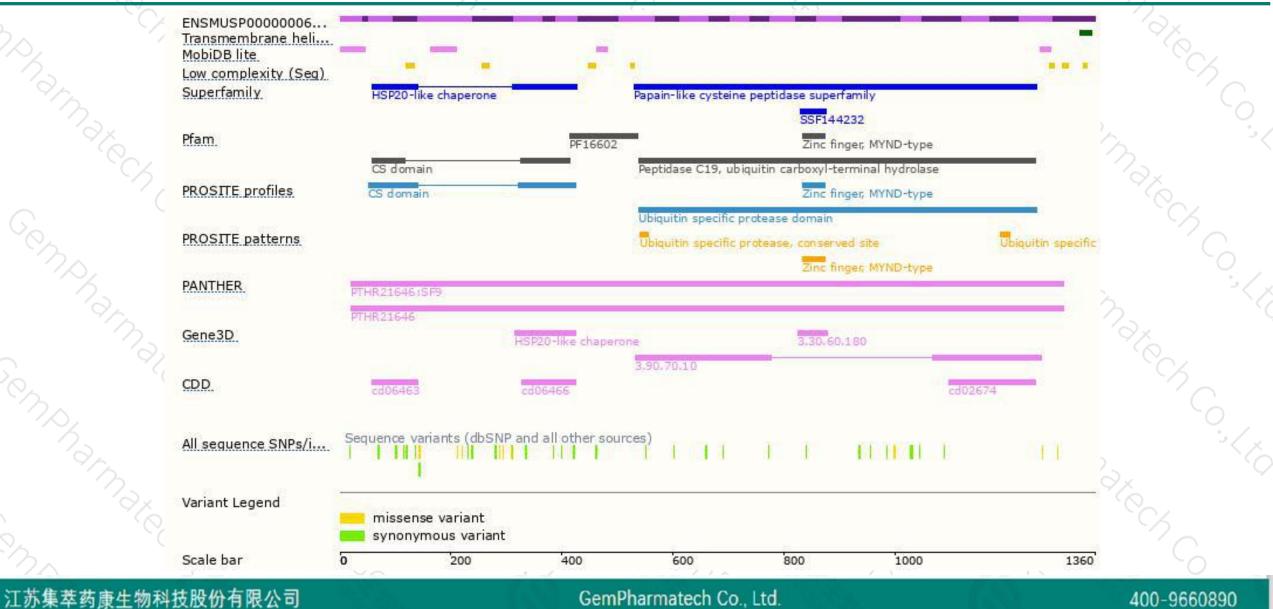


江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

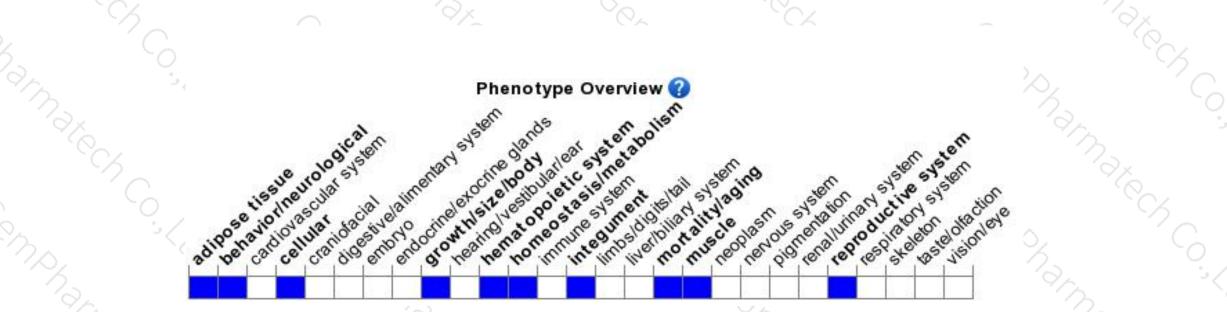
Protein domain





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 null allele exhibit decreased body weight, reduced male fertility, and increased resistance to skeletal muscle atrophy induced by both glucocorticoids and denervation.



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



