

S100a4 Cas9-KO Strategy

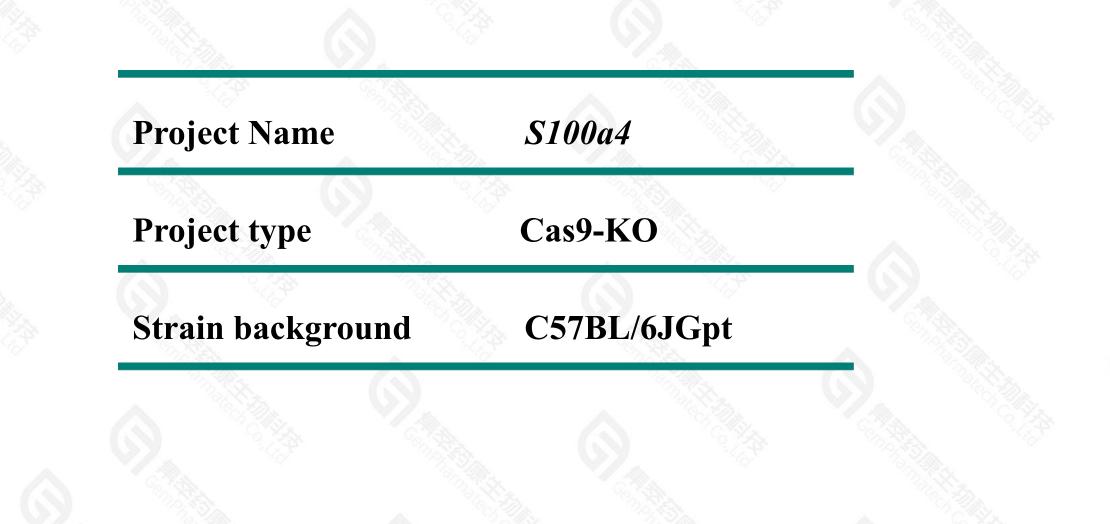
Designer: Daohua Xu

Reviewer: Huimin Su

Design Date: 2019-11-14

Project Overview





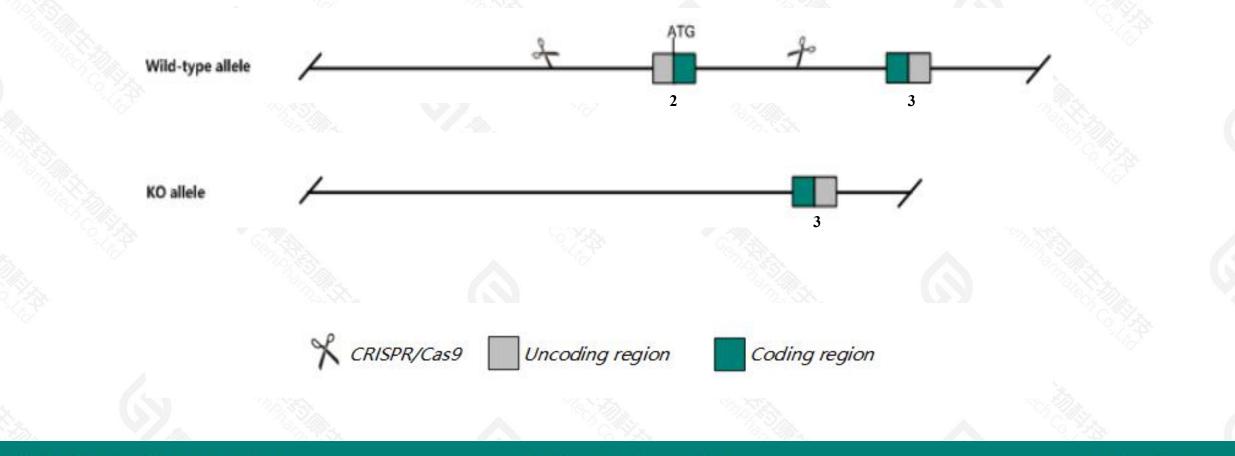
江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the S100a4 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 S100a4 gene has 2 transcripts. According to the structure of S100a4 gene, exon2 of S100a4-201(ENSMUST0000001046.7) 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 S100a4 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.



- > According to the existing MGI data, mice homozygous for one knock-out allele exhibit impaired chemotaxis of macrophages. Mice homozygous for another knock-out allele exhibit embryonic lethality resulting in a skewed sex ratio and increased tumor incidence.
- The knockout region contains the *Gm42674* gene.Knockout the region may affect the function of the *Gm42674* gene.
 The *S100a4* gene is located on the Chr3. 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.

Gene information (NCBI)



< ?

S100a4 S100 calcium binding protein A4 [Mus musculus (house mouse)]

Gene ID: 20198, updated on 13-Mar-2020

Summary

Official SymbolS100a4 provided by MGIOfficial Full NameS100 calcium binding protein A4 provided by MGIPrimary sourceMGI:MGI:1330282See relatedEnsembl:ENSMUSG0000001020Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Muroidea; Murinae; Mus; MusAlso known as18A2, 42a, Capl, FSp1, Mts1, Pe198, metastasin, pk9aExpressionBiased expression in bladder adult (RPKM 53.9), mammary gland adult (RPKM 13.8) and 10 other tissues
See more
human all

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

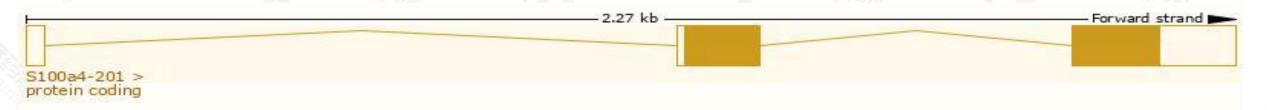
Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

Name	Transcript ID		Protein	Biotype	CCDS	UniProt	Flage
Name	Tanacripe ib	nh	Trotein	Diotype	CCDJ	Unit TOC	1 1049
S100a4-201	ENSMUST0000001046.6	498	<u>101aa</u>	Protein coding	CCDS17538	P07091 Q545V2	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
S100a4-202	ENSMUST00000142476.1	336	<u>78aa</u>	Protein coding	-	A0A0G2JGD2	CDS 3' incomplete TSL:1
225.5						0-2	

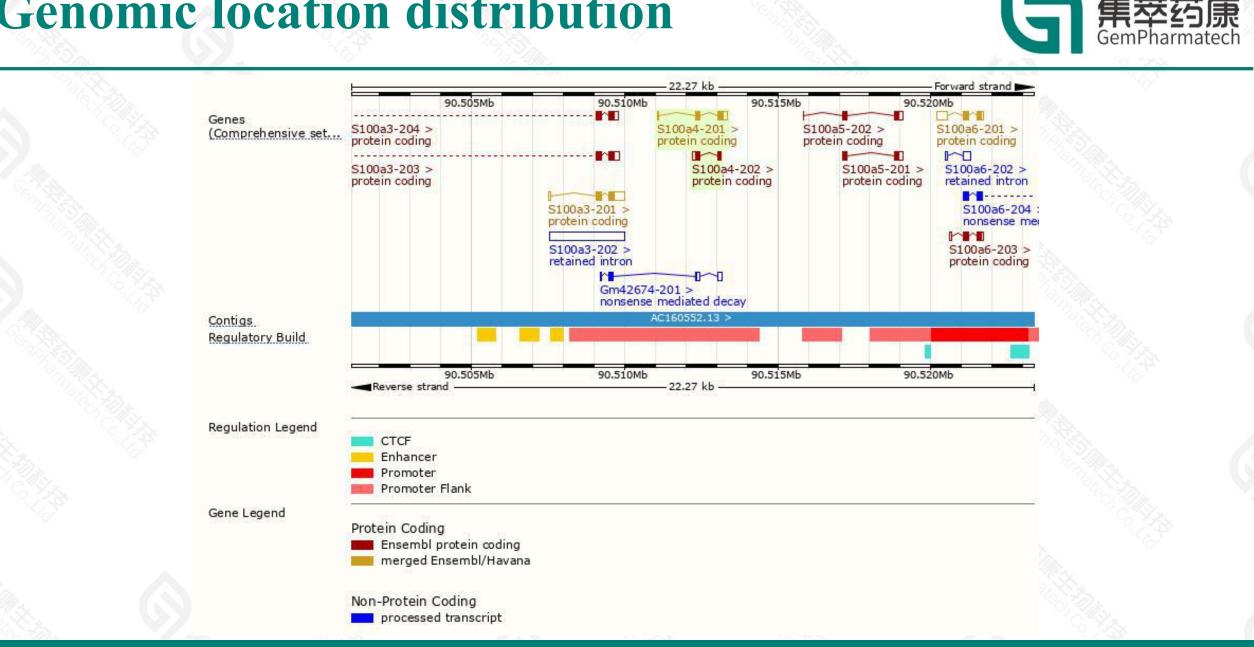
The strategy is based on the design of S100a4-201 transcript, the transcription is shown below:



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



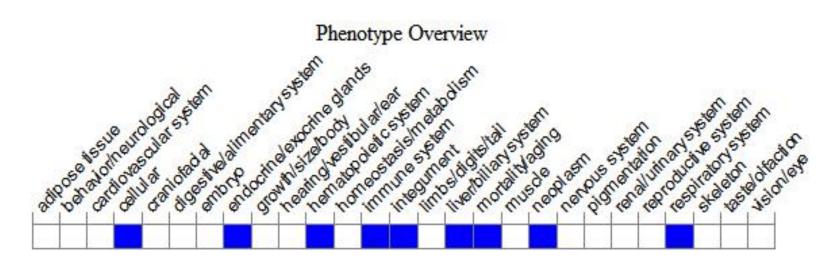
ENSMUSP00000001			-										
Superfamily	EF-hand domain	pair.											
SMART	S100/CaBP-9k	-type, calciu	um binding, su	ubdomain	EF	-hand domain	E.						
Yfam.	S100/CaBP-9k	-type, calciu	um binding, si	ubdomain									
ROSITE profiles					EF-hand	domain		3					
ROSITE patterns						EF	Hand 1, calci	um-binding si	te				
						S100/Calb	indin-D9k, co	nserved site					
ANTHER	PTHR11639									-			
	Protein S100-A4									144			
Gene3D	1.10.238.10												
DD.	S-100												
Il sequence SNPs/i	Sequence variants (dbSNP and all other sources)												
Variant Legend	missense va	riant											
Scale bar	0 10	20	30	40	50	60	70	80	90	101			

江苏集萃药康生物科技股份有限公司

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 one knock-out allele exhibit impaired chemotaxis of macrophages. Mice homozygous for another knock-out allele exhibit embryonic lethality resulting in a skewed sex ratio and increased tumor incidence.

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.



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



