

Cstfl Cas9-CKO Strategy

Designer: Reviewer:

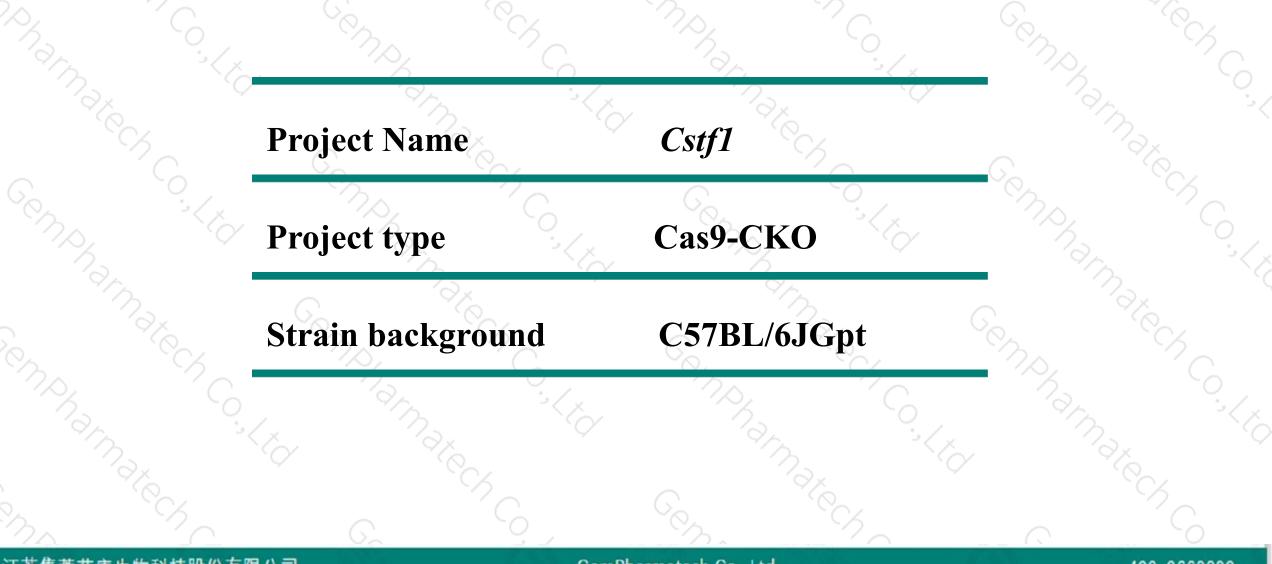
0

Design Date:

Daohua Xu Huimin Su 2019-12-17

Project Overview





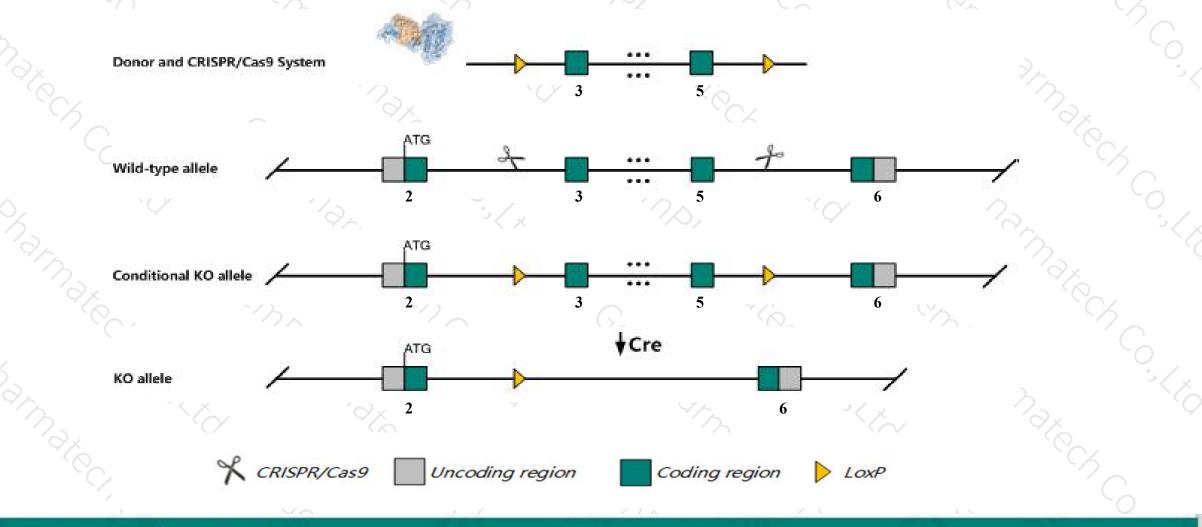
江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cstf1* 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 Cstfl gene has 2 transcripts. According to the structure of Cstfl gene, exon3-exon5 of Cstfl-201 (ENSMUST00000116375.1) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Cstf1* 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.



- The Cstfl gene is located on the Chr2. 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)



\$?

Cstf1 cleavage stimulation factor, 3' pre-RNA, subunit 1 [Mus musculus (house mouse)]

Gene ID: 67337, updated on 19-Feb-2019

Summary

Official Symbol	Cstf1 provided by MGI						
Official Full Name	cleavage stimulation factor, 3' pre-RNA, subunit 1 provided by MGI						
Primary source	MGI:MGI:1914587						
See related	Ensembl:ENSMUSG0000027498						
Gene type	protein coding						
RefSeq status	PROVISIONAL						
Organism	Mus musculus						
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;						
	Muroidea; Muridae; Murinae; Mus; Mus						
Also known as	1700057K18Rik, AI788832						
Expression	Ubiquitous expression in testis adult (RPKM 41.6), thymus adult (RPKM 23.7) and 28 other tissues See more						
Orthologs	human all						

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

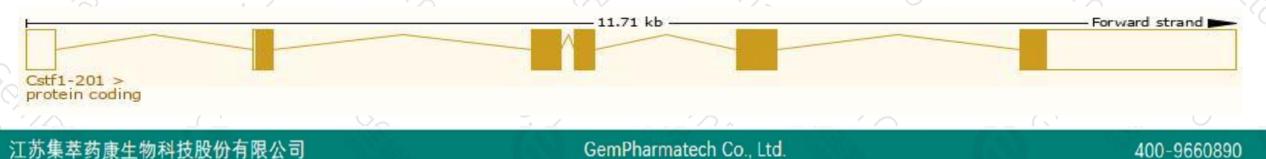
Transcript information (Ensembl)



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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cstf1-201	ENSMUST00000116375.1	3451	<u>431aa</u>	Protein coding	CCDS17130	Q99LC2	TSL:1 GENCODE basic APPRIS P1
Cstf1-202	ENSMUST00000151511.7	674	<u>155aa</u>	Protein coding	-	A2APA3	CDS 3' incomplete TSL:2

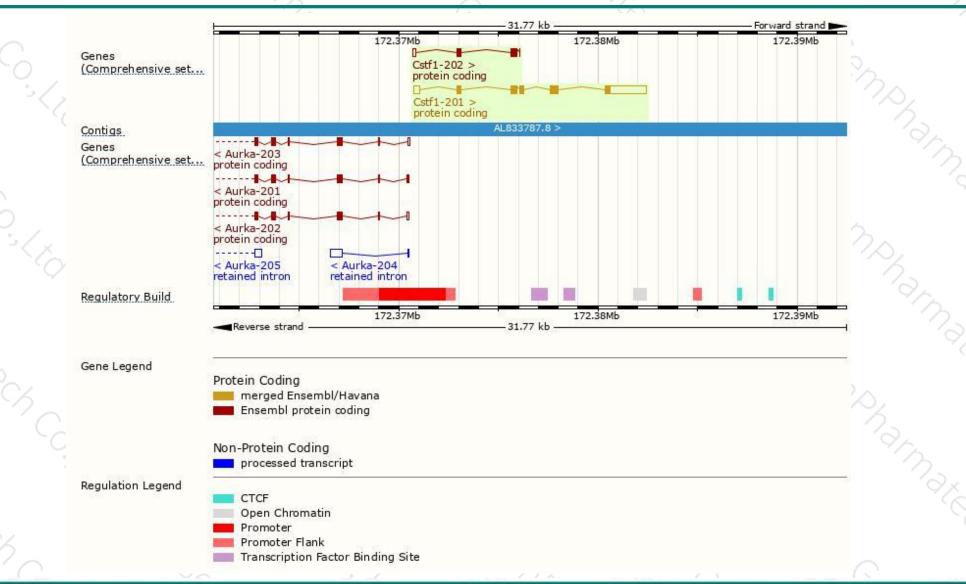
The strategy is based on the design of *Cstf1-201* transcript, The transcription is shown below



Genomic location distribution



400-9660890



江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

Protein domain



2	ENSMUSP00000112 Superfamily		WD40-repeat-o	containing domain super	family		- 70
4	SMART		WD40 repeat	-	-		
	<u>Pfam</u>	Cleavage stimulation :		risation domain			م
			WD40 repeat				
	PROSITE profiles		provide the second s	containing domain			
			WD40 repeat				
	PROSITE patterns					WD40 repeat, conserved	d site
	PANTHER	PTHR44133					
	Gene3D		WD40/YVTN.re	peat-like-containing dor	nain superfamily		
		CSTF1, dimerization dor	nain superfamily				
	CDD	- college trevelation of the	cd00200				
	All sequence SNPs/i	Sequence variants (c	bSNP and all other :	sources)	67. 3	1 12 1	6
	Variant Legend						
		— missense variar — synonymous va					
	Scale bar	o 40	80 120	160 200	240 280	320 360	431
		v 10		100 200	240 200	520 500	
			~ 0		1		

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



