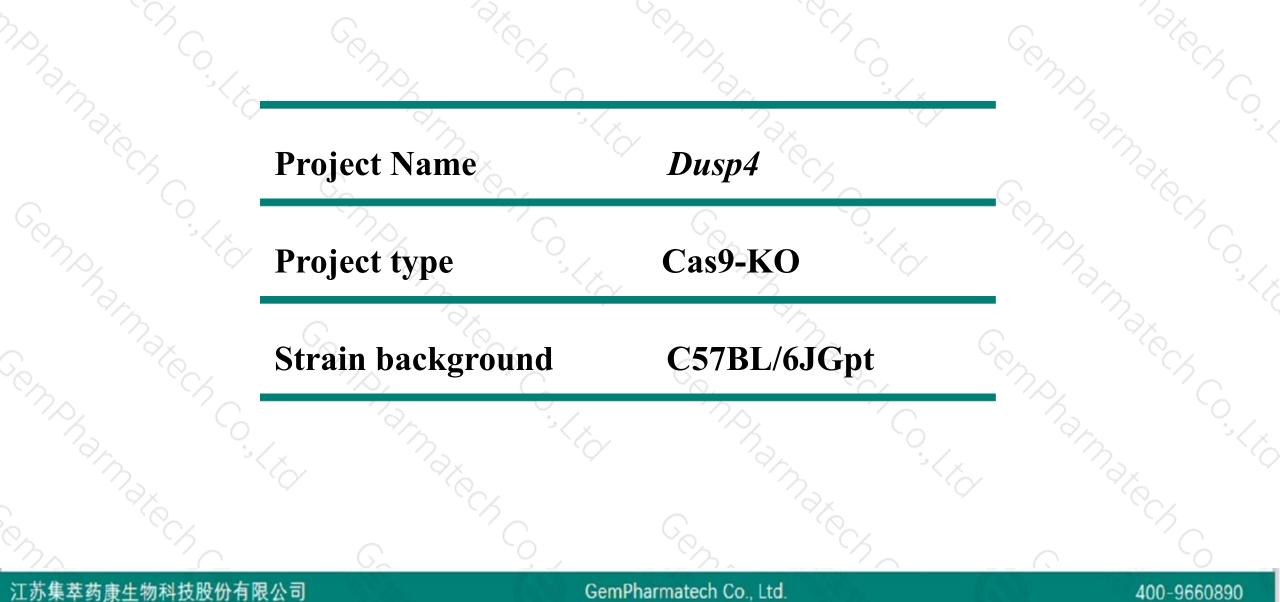


Dusp4 Cas9-KO Strategy

Designer: Reviewer: Design Date: Ruirui Zhang Huimin Su 2020-2-28

Project Overview

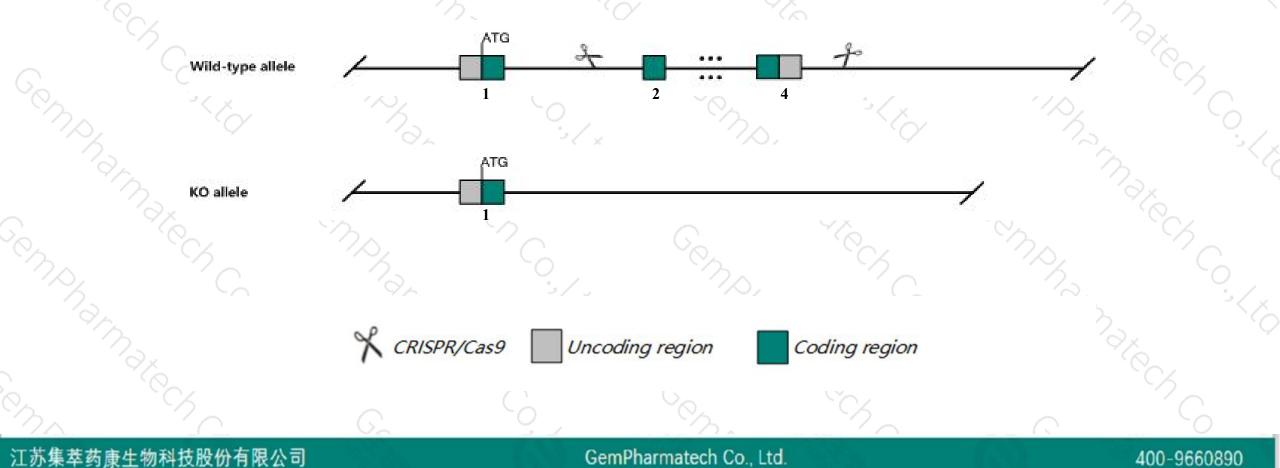




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Dusp4* gene. The schematic diagram is as follows:





- The Dusp4 gene has 1 transcript. According to the structure of Dusp4 gene, exon2-exon4 of Dusp4-201 (ENSMUST00000033930.4) transcript is recommended as the knockout region. The region contains most of coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Dusp4* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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 a null allele exhibit a decrease in B cell apoptosis of bone marrow-derived, IL-7-dependent pro-B lymphocytes.
- The Dusp4 gene is located on the Chr8. 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.

Notice

Gene information (NCBI)



Dusp4 dual specificity phosphatase 4 [Mus musculus (house mouse)]

Gene ID: 319520, updated on 31-Dec-2019

Summary

Official Symbol	Dusp4 provided by MGI								
Official Full Name	dual specificity phosphatase 4 provided by MGI								
Primary source	MGI:MGI:2442191								
See related	Ensembl:ENSMUSG00000031530								
Gene type	protein coding								
RefSeq status	PROVISIONAL								
Organism	<u>Mus musculus</u>								
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae;								
	Murinae; Mus; Mus								
Also known as	MKP2; AI844617; BB104621; 2700078F24Rik; E130306H24Rik								
Expression	Broad expression in whole brain E14.5 (RPKM 27.6), CNS E14 (RPKM 24.4) and 20 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.

400-9660890

☆ ?

Transcript information (Ensembl)



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

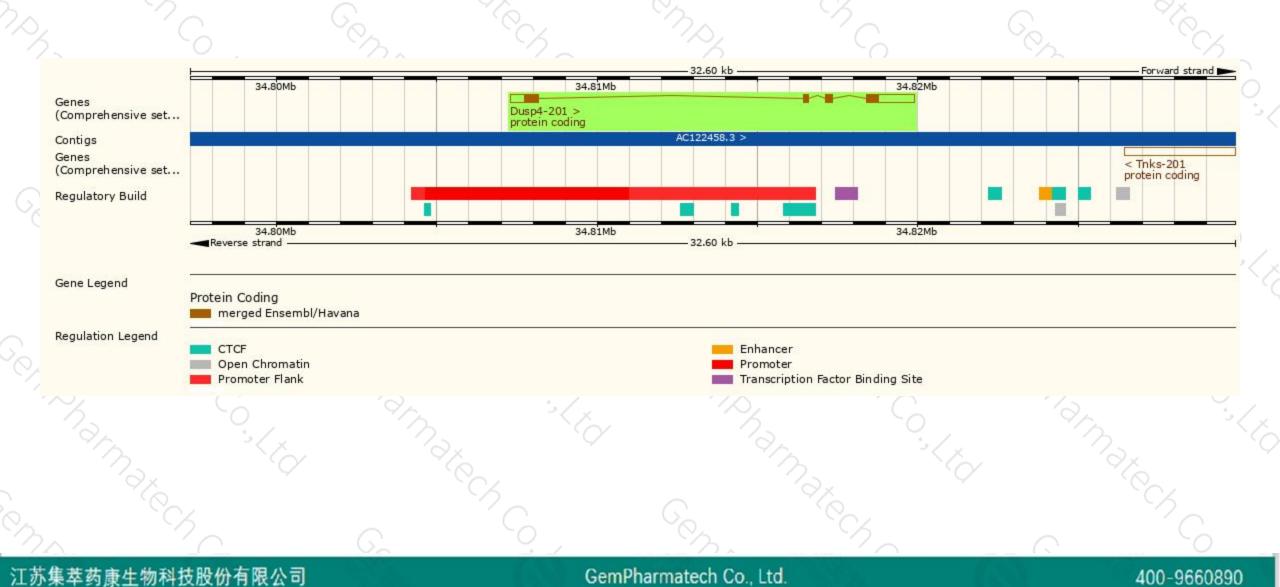
Name 🖕	Transcript ID 🖕	bp 🖕	Protein 🖕	Biotype 🔺	CCDS 🖕	UniProt 🖕		Flags	
Dusp4-201	ENSMUST0000033930.4	2740	<u>398aa</u>	Protein coding	<u>CCDS22241</u> &	<u>Q8BFV3</u> &	TSL:1	GENCODE basic	APPRIS P1

The strategy is based on the design of *Dusp4-201* transcript, the transcription is shown below:



Genomic location distribution





Protein domain



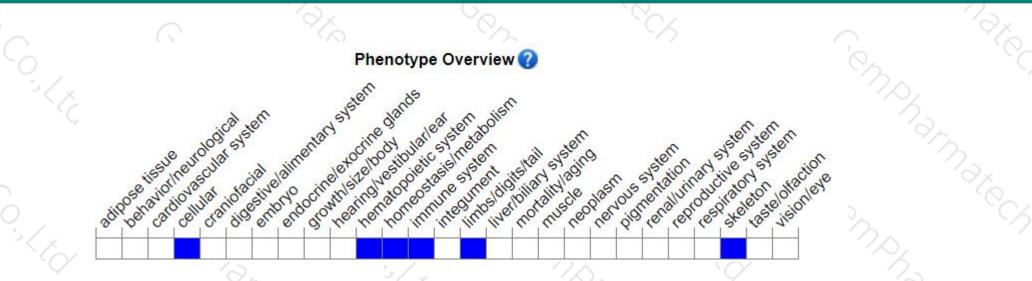
5									
ENSMUSP00000033 Low complexity (Seg)									
Superfamily	Rhodanese-like domain superfamily			Protein-tyrosine ph	osphatase-like				
SMART	Rhodanese-like domain				102-25-510-27-27-25-25-25-25-25-25-25-25-25-25-25-25-25-	e phosphatase, cata	lytic		0
Prints	Mitogen-activated proto	n (MAP) kinase phosphatase		Dual specifici	ty protein phosphatase d	omain			
Pfam	Rhodanese-like domain	n (mor) kinase prosphatase	-	Dual spe	cificity phosphatase, cata	lytic domain			
PROSITE profiles	Rhodanese-like domain		1	Dual specifici	Tr		ein phosphatases domain		
PROSITE patterns				buar specific	ry process prosphacase o	Contraction and the second second	ine phosphatase, active sit	te	
PIRSF	Mitogen-activated protein (MAP) kinase phosphata	se						14	➡,∠
PANTHER	PTHR10159:SF111								
Gene3D	PTHR10159 Rhodanese-like domain superfamily			Protein-tyrosir	e phosphatase-like				
CDD	cd01446		_	cd14640					
All sequence SNPs/i	Sequence variants (dbSNP and all other source	es)	r -	100 - O	n i i	- a -	10 D.	а.	uk _é
Variant Legend	inframe deletion stop retained variant				nse variant ymous variant				
Scale bar	o 40 80	120	160	200	240	280	320		398
'A'				×>.	~~~~				
			and the second		the second se				

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

400-9660890

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 a decrease in B cell apoptosis of bone marrow-derived, IL-7-dependent pro-B lymphocytes.

江苏集萃药康生物科技股份有限公司

GemPharmatech Co., Ltd.

400-9660890



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



