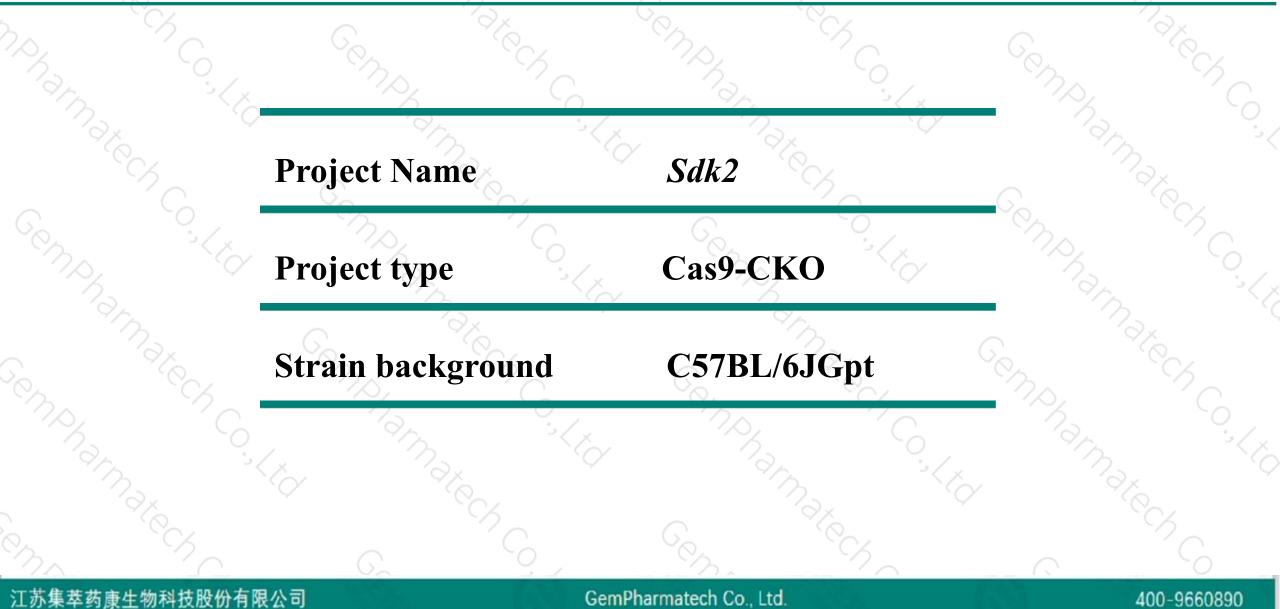


Sdk2 Cas9-CKO Strategy

Designer: Reviewer: Design Date: Daohua Xu Huimin Su 2020-3-30

Project Overview

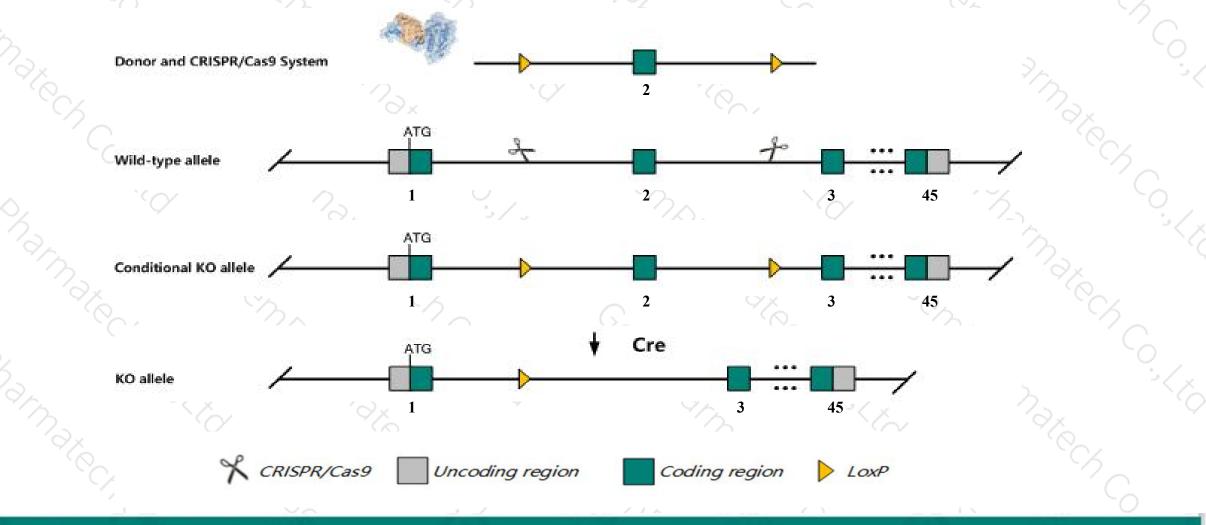




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Sdk2* gene. The schematic diagram is as follows:



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 The Sdk2 gene has 6 transcripts. According to the structure of Sdk2 gene, exon2 of Sdk2-201 (ENSMUST00000041627.13) transcript is recommended as the knockout region. The region contains 160bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Sdk2* 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 knock-out allele exhibit impaired interconnectvity between VG3 amacrine cells and W3B retinal ganglion cells.
 - > The Sdk2 gene is located on the Chr11. 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)



\$?

Sdk2 sidekick cell adhesion molecule 2 [Mus musculus (house mouse)]

Gene ID: 237979, updated on 13-Mar-2020

Summary

Official Symbol	Sdk2 provided by MGI								
Official Full Name	sidekick cell adhesion molecule 2 provided by MGI								
Primary source	MGI:MGI:2443847								
See related	Ensembl:ENSMUSG00000041592								
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	4632412F08Rik, 5330435L01Rik, Sdk-2, mKIAA1514								
Expression	Broad expression in limb E14.5 (RPKM 3.2), frontal lobe adult (RPKM 1.8) and 22 other tissues See more								
Orthologs	human all								

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Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
0.07500.05	ENSMUST00000041627.13	0.00		Protein coding			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
5dk2-203	ENSMUST00000141943.1	2783	<u>895aa</u>	Protein coding	-	B1ASR7	CDS 3' incomplete TSL:1
3dk2-205	ENSMUST00000155651.1	4155	No protein	Processed transcript		0.20	TSL1
6dk2-204	ENSMUST00000144301.1	3083	No protein	Processed transcript	-	1926	TSL:1
dk2-202	ENSMUST00000125879.1	681	No protein	Processed transcript		1070	TSL:3
dk2-206	ENSMUST00000156955.1	546	No protein	Processed transcript			TSL:3
JUK2-200	ENSINGS100000130333.1	540	No protein	1 rocessed transcript	1		

The strategy is based on the design of *Sdk2-201* transcript, The transcription is shown below

< Sdk2-201 protein coding

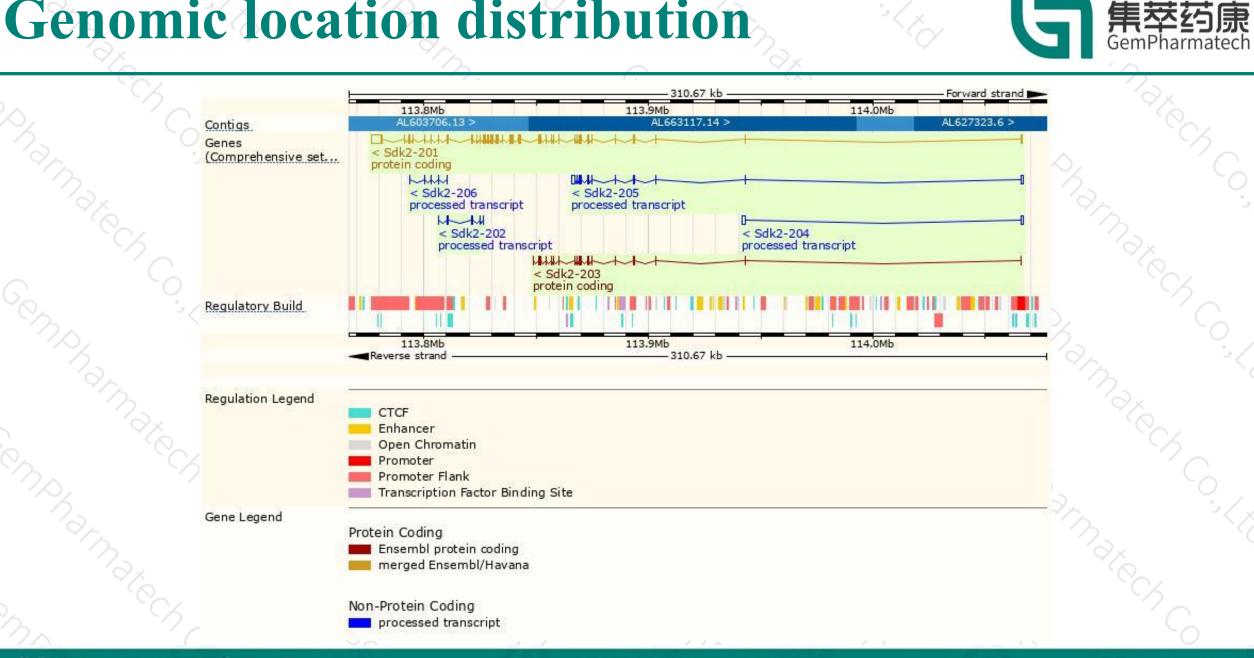
Reverse strand

_____ 289.85 kb

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Genomic location distribution



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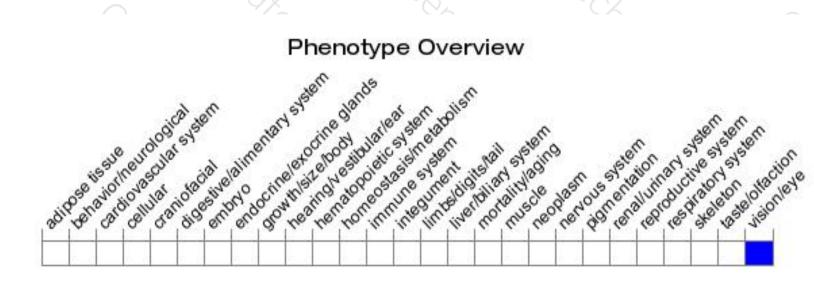
Protein domain



	PDB-ENSP mappings MobiDB lite		- ~~
	Low complexity (Seg) Cleavage site (Sign		
- 19K	Superfamily	Immunoglobulin-like domain superfamily	
	SMART	Fibronectin type III superfamily Immunoglobulin subtype 2 Fibronectin type III	
	Prints	Immunoglobulin subtype PR00014	° CA
	Pfam.	Immunoglobulin I-set Fibronectin type III	
	PROSITE profiles PANTHER	PF13927 Immunoglobulin-like domain Fibronectin type III PTHR13817:SF59	
	Gene3D CDD	PTHR13817 Immunoglobulin-like fold cd00096 Fibronectin type III	°C /
non and a second	All sequence SNPs/i	Sequence variants (dbSNP and all other sources)	· · · · · · · · · · · · · · · · · · ·
ALL ALL	Variant Legend	missense variant splice region variant	
		synonymous variant	
	Scale bar	0 200 400 600 800 1000 1200 1400 1600 1800	2176

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 knock-out allele exhibit impaired interconnectvity between VG3 amacrine cells and W3B retinal ganglion cells.



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



