

Dcaf12 Cas9-CKO Strategy

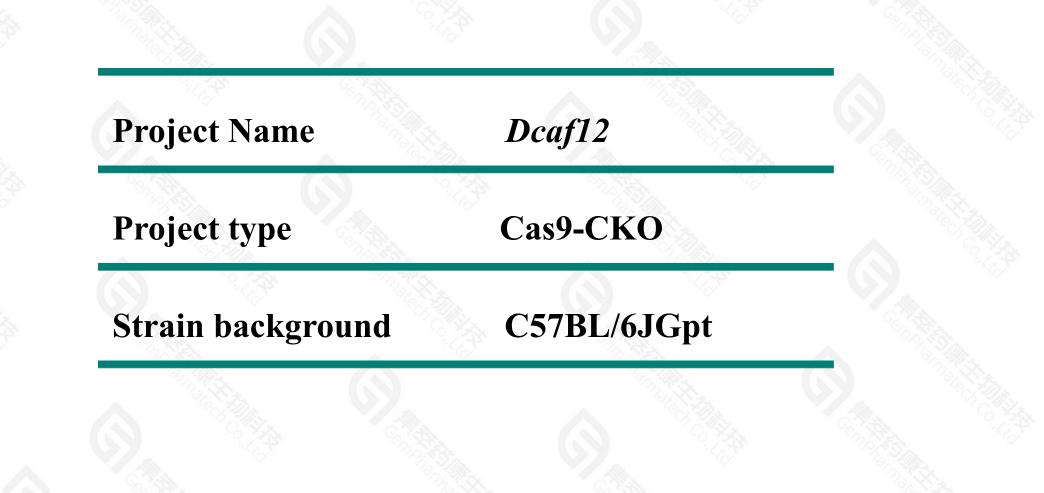
Designer: Huan Wang

Reviewer: Yumeng Wang

Design Date: 2021-9-27

Project Overview





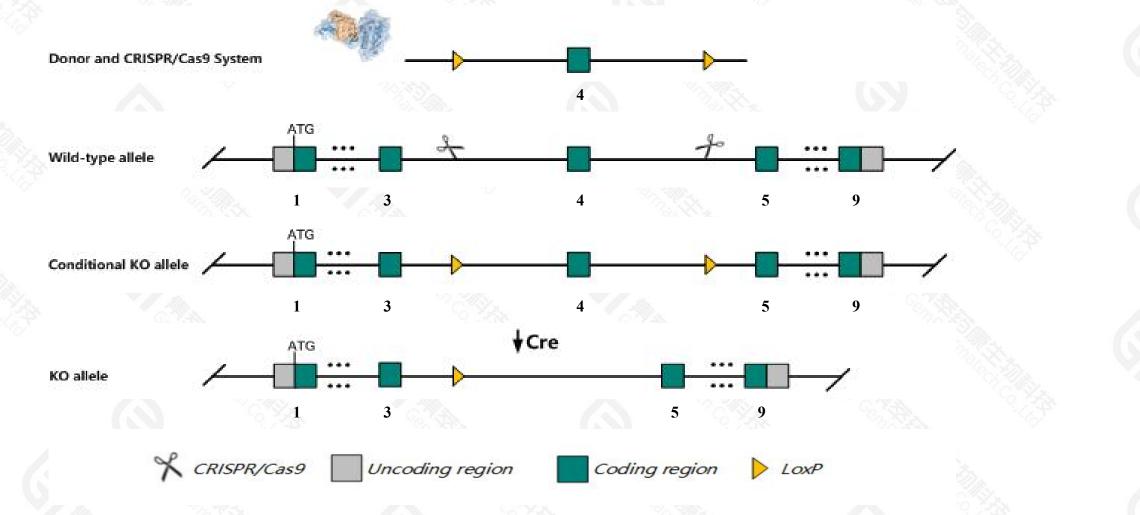
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Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Dcaf12* gene. The schematic diagram is as follows:



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Technical routes



The *Dcaf12* gene has 1 transcript. According to the structure of *Dcaf12* gene, exon4 of *Dcaf12-201*(ENSMUST00000030145.9) transcript is recommended as the knockout region. The region contains 61bp coding sequence. Knock out the region will result in disruption of protein function.

> In this project we use CRISPR/Cas9 technology to modify *Dcaf12* 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



- > The *Dcaf12* gene is located on the Chr4. 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.
- > Some amino acids remain at the N terminal, and part of the protein's function may be preserved.
- > 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.

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Gene information (NCBI)

Dcaf12 DDB1 and CUL4 associated factor 12 [Mus musculus (house mouse)]

Gene ID: 68970, updated on 17-Dec-2020

Summary

Official Symbol	Dcaf12 provided by MGI
Official Full Name	DDB1 and CUL4 associated factor 12 provided by MGI
Primary source	MGI:MGI:1916220
See related	Ensembl:ENSMUSG0000028436
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	1500001L20Rik, 5830424K06Rik, AA420338, AI851081, Wdr4, Wdr40a
Expression	Ubiquitous expression in testis adult (RPKM 28.9), liver E14 (RPKM 16.7) and 28 other tissues See more
Orthologs	human all



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Transcript information (Ensembl)

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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dcaf12-201	ENSMUST0000030145.9	3442	<u>453aa</u>	Protein coding	CCDS18059		TSL:1, GENCODE basic, APPRIS P1,

The strategy is based on the design of *Dcaf12-201* transcript, the transcription is shown below:

< Dcaf12-201 protein coding

Reverse strand

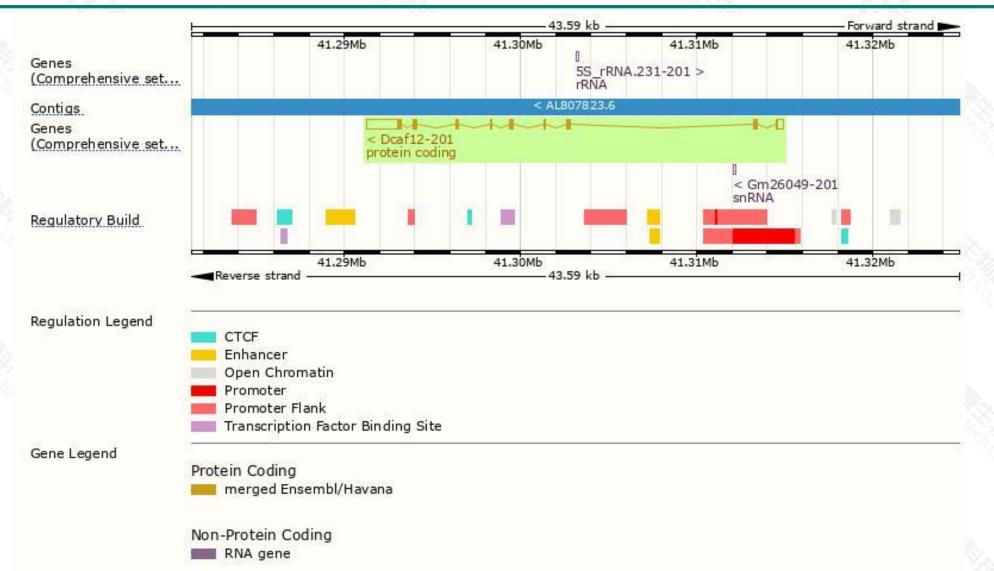
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Genomic location distribution





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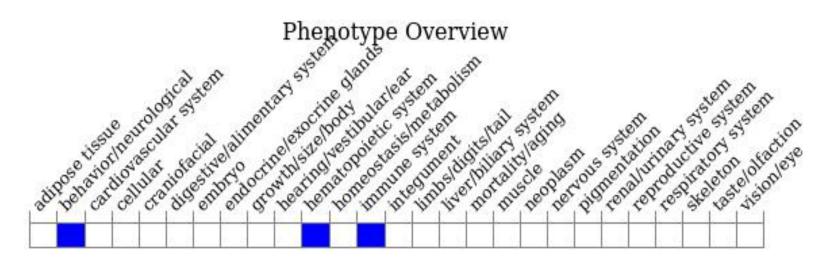
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Protein domain



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SMART		WD40 r	epeat	-		-	-	-			
PROSITE profiles				W	D40 repeat						
				W	D40-repeat	containin	g domain				
PROSITE patterns							served site				
PANTHER	PTHR19860					<u>.</u>					-
	FTHR 19860 :SF	10									
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Gene3D		WD40/YVTN re	peat-like-cor	ntaining dom	ain superfa	mily					
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All sequence SNPs/i	Sequence va	riants (dbSNP				mily	Ŀя	л	A)	r	
Gene3D All sequence SNPs/i Variant Legend	Sequence va					mily	E I		Ľ	T	

Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).



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



