Dusp8 Cas9-CKO Strategy

Designer:

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Design Date:

2019-9-29

Project Overview



Project Name

Dusp8

Project type

Cas9-CKO

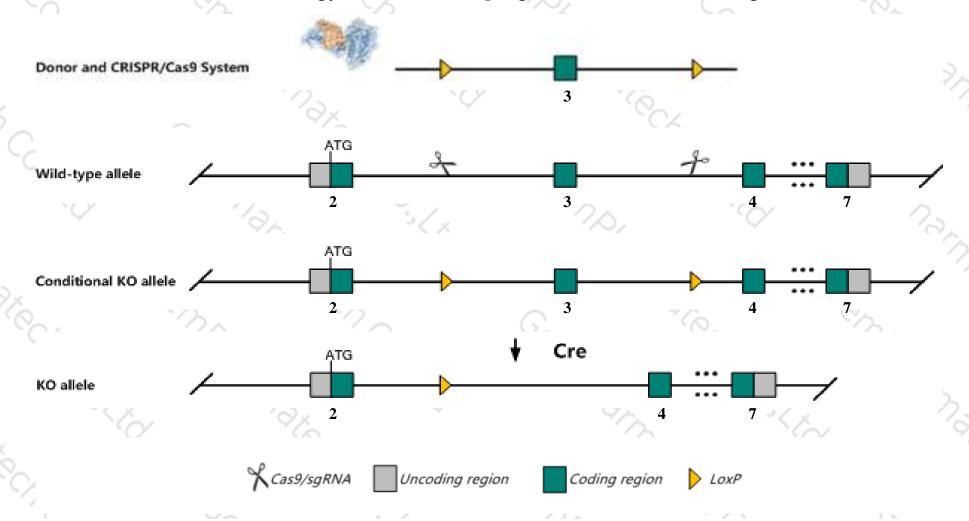
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Dusp8* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Dusp8* gene has 2 transcripts. According to the structure of *Dusp8* gene, exon3 of *Dusp8-201*(ENSMUST00000039926.9) transcript is recommended as the knockout region. The region contains 139bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Dusp8* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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.

Notice



- ➤ According to the existing MGI data, Mice homozygous for a knock-out allele exhibit altered myocardial fiber morphology, mildly increased cardiac muscle contractility at baseline, and decreased response of heart to induced stress.
- ➤ The *Dusp8* gene is located on the Chr7. 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)



Dusp8 dual specificity phosphatase 8 [Mus musculus (house mouse)]

Gene ID: 18218, updated on 12-Aug-2019

Summary

☆ ?

Official Symbol Dusp8 provided by MGI

Official Full Name dual specificity phosphatase 8 provided by MGI

Primary source MGI:MGI:106626

See related Ensembl:ENSMUSG00000037887

Gene type protein coding
RefSeq status VALIDATED
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 Nttp1; Al593498; 5530400B01Rik

Expression Broad expression in whole brain E14.5 (RPKM 20.5), CNS E18 (RPKM 19.5) and 22 other tissues See more

Orthologs <u>human</u> <u>all</u>

▲ Genomic context

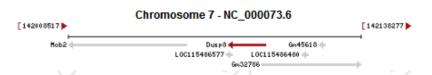


Location: 7 F5; 7 87.59 cM

See Dusp8 in Genome Data Viewer

Exon count: 8

Annotation release	Status	Assembly	Chr	Location	
<u>108</u>	current	GRCm38.p6 (GCF 000001635.26)	7	NC_000073.6 (142079487142095907, complement)	
Build 37.2	previous assembly	MGSCv37 (GCF 000001635.18)	7	NC_000073.5 (149267401149276175, complement)	



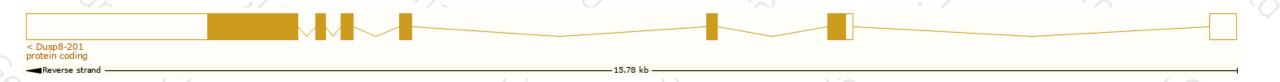
Transcript information (Ensembl)



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

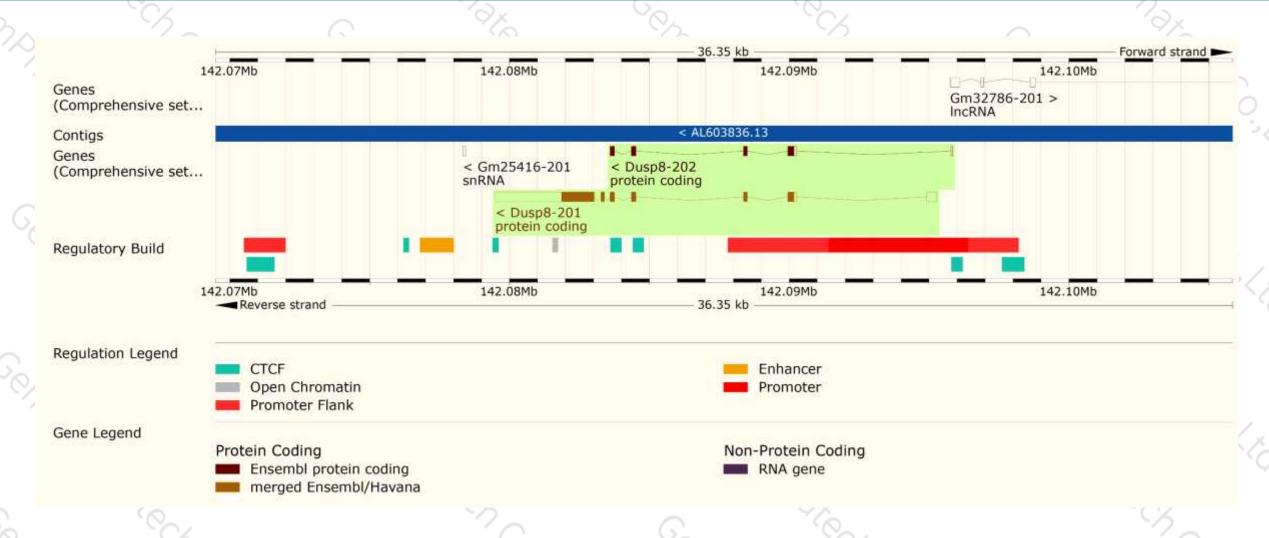
Show/hid	e columns (1 hidden)	Filter					
Name 🍦	Transcript ID	bp 🌲	Protein	Biotype	CCDS 🍦	UniProt 🍦	Flags
Dusp8-201	ENSMUST00000039926.9	4804	<u>663aa</u>	Protein coding	CCDS22024 ₽	<u>009112</u> 配	TSL:1 GENCODE basic APPRIS P1
Dusp8-202	ENSMUST00000143661.1	853	<u>231aa</u>	Protein coding	-	<u>B7ZC52</u> ₽	CDS 3' incomplete TSL:3

The strategy is based on the design of *Dusp8* -201 transcript, The transcription is shown below



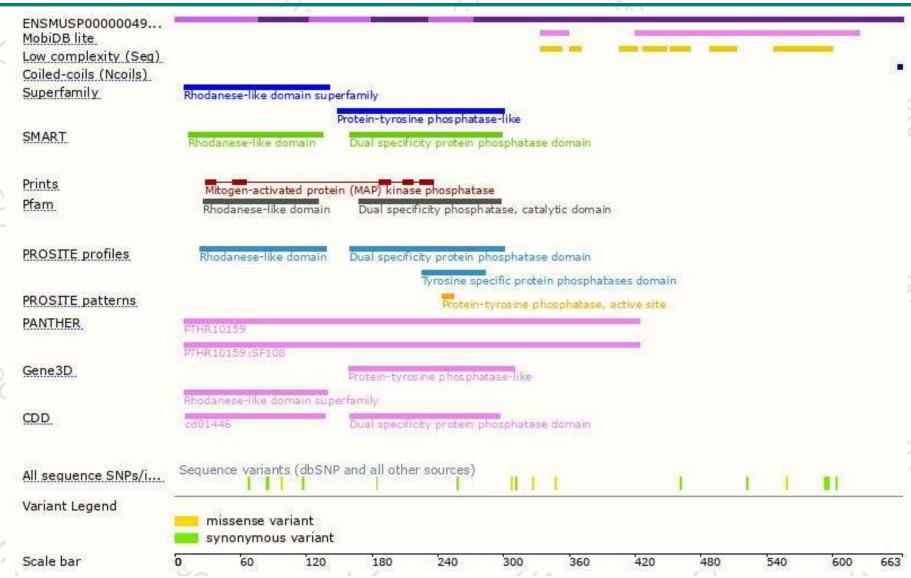
Genomic location distribution





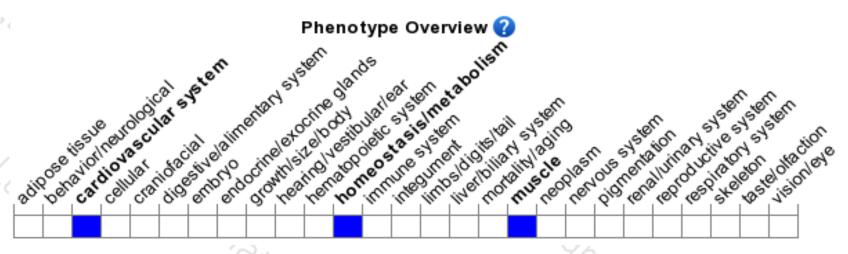
Protein domain





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 altered myocardial fiber morphology, mildly increased cardiac muscle contractility at baseline, and decreased response of heart to induced stress.

If you have any questions, you are welcome to inquire. Tel: 025-5864 1534





