

Dlc1 Cas9-KO Strategy

Designer: JiaYu

Reviewer: Xiaojing Li

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Project Overview



Project Name Dlc1

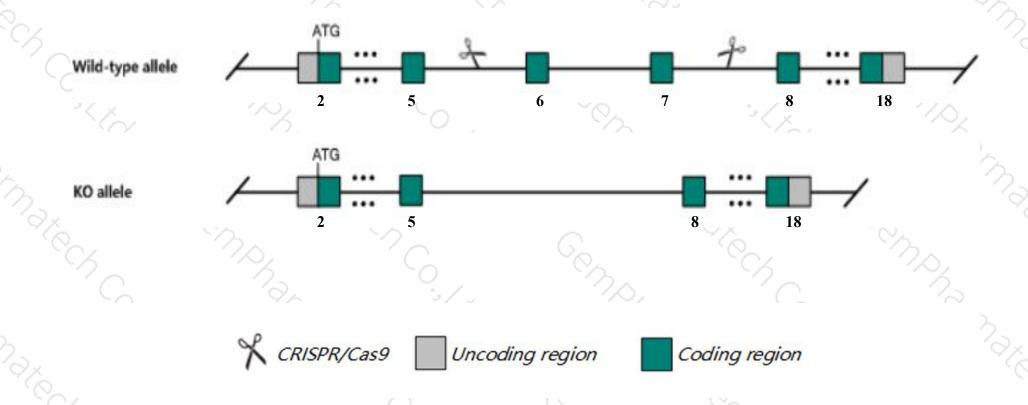
Project type Cas9-KO

Strain background C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Dlc1* gene. The schematic diagram is as follows:



Technical routes



- The *Dlc1* gene has 9 transcripts. According to the structure of *Dlc1* gene, exon6-exon7 of *Dlc1*206(ENSMUST00000163663.2) transcript is recommended as the knockout region. The region contains 154bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Dlc1* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice



- > According to the existing MGI data, homozygous mutants die by E10.5 with variable defects in the neural tube, heart, brain and placenta. Mouse embryonic fibroblasts homozygous for an activated conditional allele exhibti increased sensitivity to Ras-induced transformation.
- ➤ Some amino acids will remain at the N-terminus and some functions may be retained.
- ➤ Transcripts 209 maybe unaffected.
- ➤ Transcript 208 CDS 3' incomplete the influences is unknown.
- The *Dlc1* 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.

Gene information (NCBI)



Dlc1 deleted in liver cancer 1 [Mus musculus (house mouse)]

Gene ID: 50768, updated on 13-Mar-2020





Official Symbol Dlc1 provided by MGI

Official Full Name deleted in liver cancer 1 provided byMGI

Primary source MGI:MGI:1354949

See related Ensembl: ENSMUSG000000031523

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 A730069N07Rik, Arhgap7, HP, STARD12, dlc-1

Expression Broad expression in lung adult (RPKM 9.5), subcutaneous fat pad adult (RPKM 8.0) and 24 other tissuesSee more

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

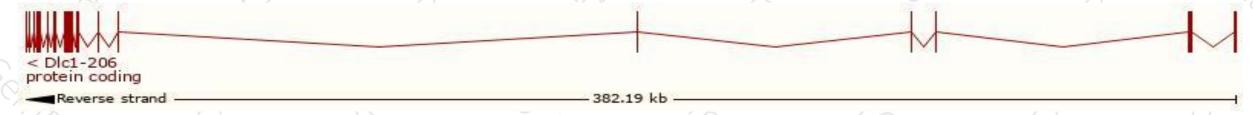
Transcript information (Ensembl)



The gene has 9 transcripts, all transcripts are shown below:

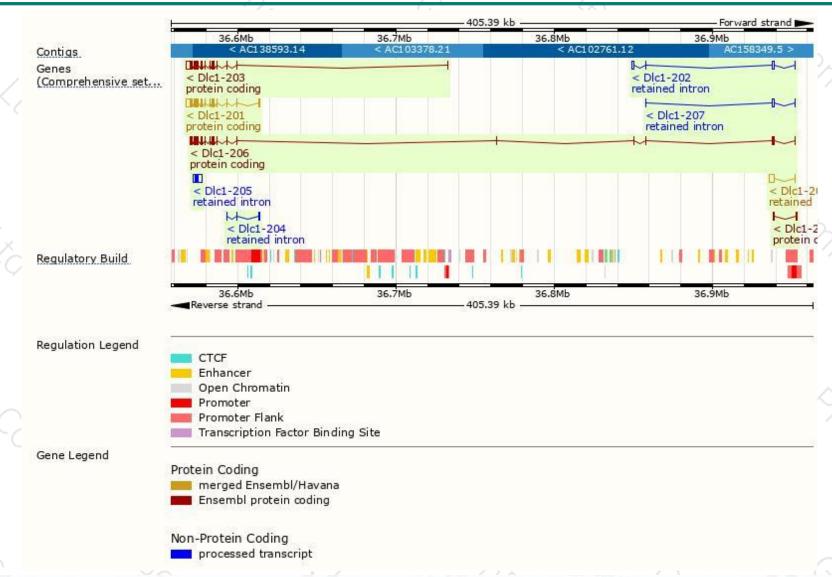
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dlc1-203	ENSMUST00000098826.9	6241	1126aa	Protein coding	CCDS57616	A0A0R4J171	TSL:1 GENCODE basic
Dlc1-201	ENSMUST00000033923.13	6159	<u>1092aa</u>	Protein coding	CCDS40322	E9QKB1	TSL:1 GENCODE basic APPRIS P1
Dlc1-206	ENSMUST00000163663.2	5113	<u>1543aa</u>	Protein coding	CCDS57617	E9PXD2	TSL:5 GENCODE basic
Dlc1-208	ENSMUST00000179501.1	403	<u>26aa</u>	Protein coding	7	J3QPV2	CDS 3' incomplete TSL:3
Dlc1-209	ENSMUST00000179652.1	3491	No protein	Retained intron	2	-	TSL:2
Dlc1-205	ENSMUST00000156312.1	3423	No protein	Retained intron	- 60	-	TSL:2
Dlc1-202	ENSMUST00000036104.10	2756	No protein	Retained intron	-	-	TSL:1
Dlc1-207	ENSMUST00000178717.1	1866	No protein	Retained intron	2	-	TSL:1
Dlc1-204	ENSMUST00000145245.1	616	No protein	Retained intron	-	-	TSL:1
	- / / /	-/-/-			N. 10 mm		A Vice

The strategy is based on the design of *Dlc1-206* transcript, the transcription is shown below:



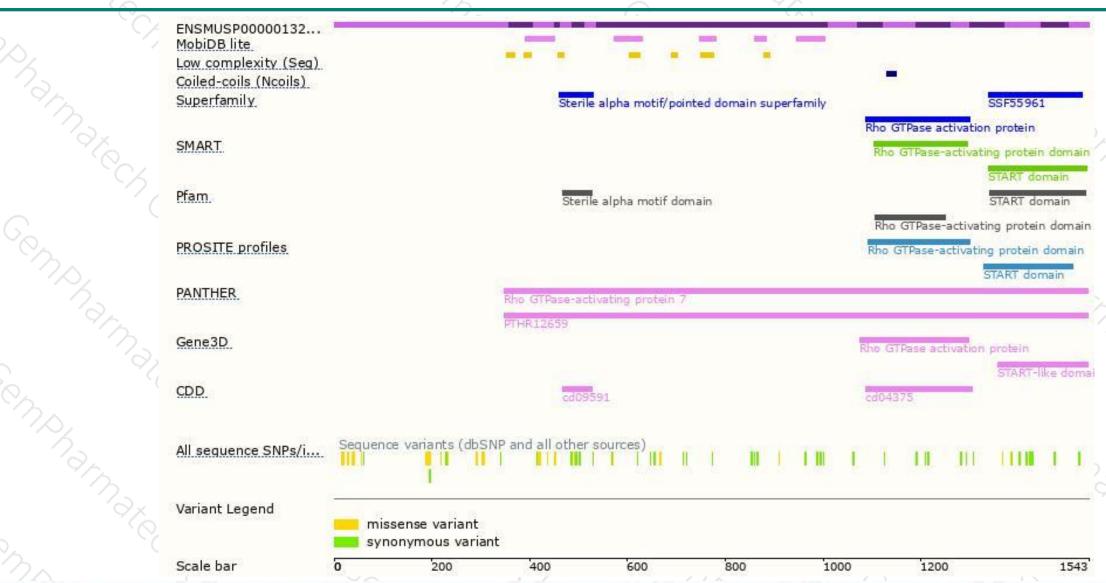
Genomic location distribution





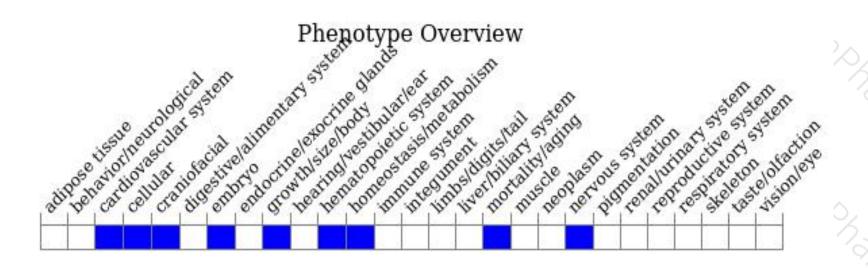
Protein domain





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, homozygous mutants die by E10.5 with variable defects in the neural tube, heart, brain and placenta. Mouse embryonic fibroblasts homozygous for an activated conditional allele exhibti increased sensitivity to Ras-induced transformation.



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





