

Dcc Cas9-CKO Strategy

Designer: Huimin Su

Reviewer: Ruirui Zhang

Design Date: 2019/12/18

Project Overview



Project Name

Project type

Strain background

Q,

Dcc

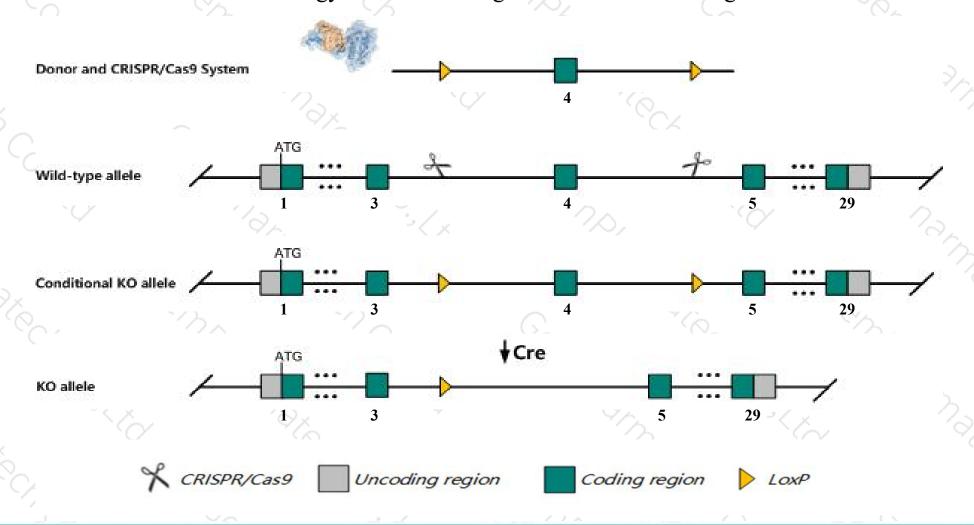
Cas9-CKO

C57BL/6JGpt

Conditional Knockout strategy



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



Technical routes



- The *Dcc* gene has 3 transcripts. According to the structure of *Dcc* gene, exon4 of *Dcc-202*(ENSMUST00000114943.10) transcript is recommended as the knockout region. The region contains 151bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Dcc* 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.

Notice



- ➤ According to the existing MGI data, Homozygous animals show defects in axonal projections and hypothalamic development affecting both visual and neruoendocrine systems. Incidence of tumors increases in mutations preventing netrin-1 binding.
- > The *Dcc* gene is located on the Chr18. 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)



Dcc deleted in colorectal carcinoma [Mus musculus (house mouse)]

Gene ID: 13176, updated on 4-Dec-2019

Summary

Official Symbol Dcc provided by MGI

Official Full Name deleted in colorectal carcinoma provided by MGI

Primary source MGI:MGI:94869

See related Ensembl: ENSMUSG00000060534

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 Igdcc1; C030036D22Rik

Expression Biased expression in whole brain E14.5 (RPKM 7.6), CNS E14 (RPKM 7.4) and 5 other tissues See more

Orthologs human all

Genomic context

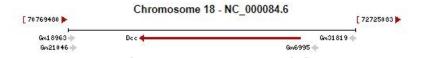
Location: 18 E2; 18 45.24 cM

See Dcc in Genome Data Viewer

☆ ?

Exon count: 29

Annotation release	Status	Assembly	Chr	Location
<u>108</u>	current	GRCm38.p6 (GCF_000001635.26)	18	NC_000084.6 (7125361372351228, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	18	NC_000084.5 (7141328672510723, complement)



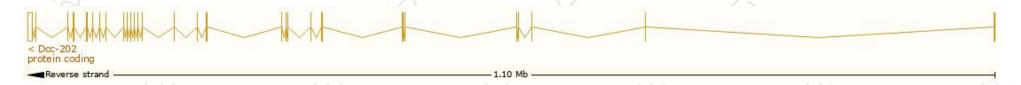
Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

Name 🍦	Transcript ID	bp 🍦	Protein	Biotype	CCDS	UniProt	Flags
Dcc-202	ENSMUST00000114943.10	10323	<u>1447aa</u>	Protein coding	CCDS29336 €	P70211 ₽	TSL:1 GENCODE basic APPRIS P1
Dcc-201	ENSMUST00000073379.5	4855	1427aa	Protein coding	-	P70211 ₽	TSL:5 GENCODE basic
Dcc-203	ENSMUST00000126030.1	3855	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Dcc-202* transcript, The transcription is shown below



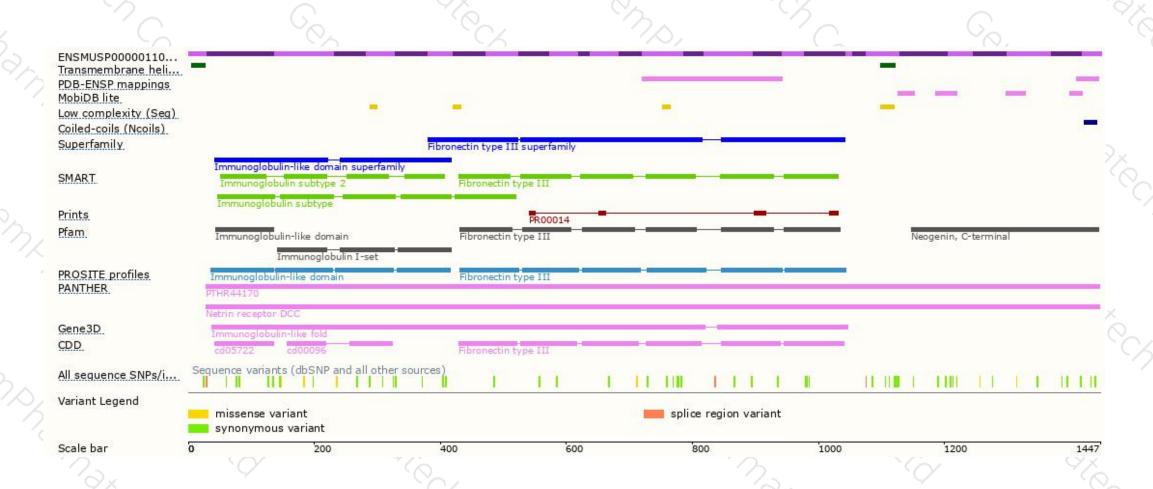
Genomic location distribution





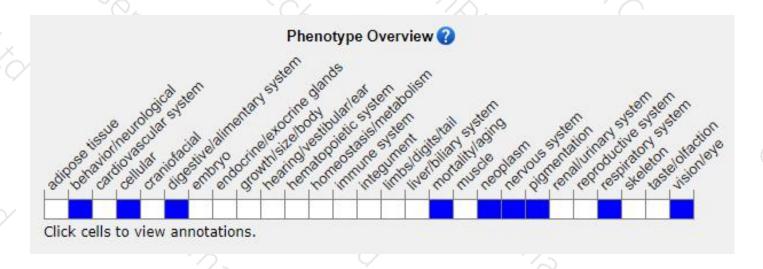
Protein domain





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 animals show defects in axonal projections and hypothalamic development affecting both visual and neruoendocrine systems. Incidence of tumors increases in mutations preventing netrin-1 binding.



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





