

Vcan Cas9-CKO Strategy

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Project Overview



Project Name

Vcan

Project type

Cas9-CKO

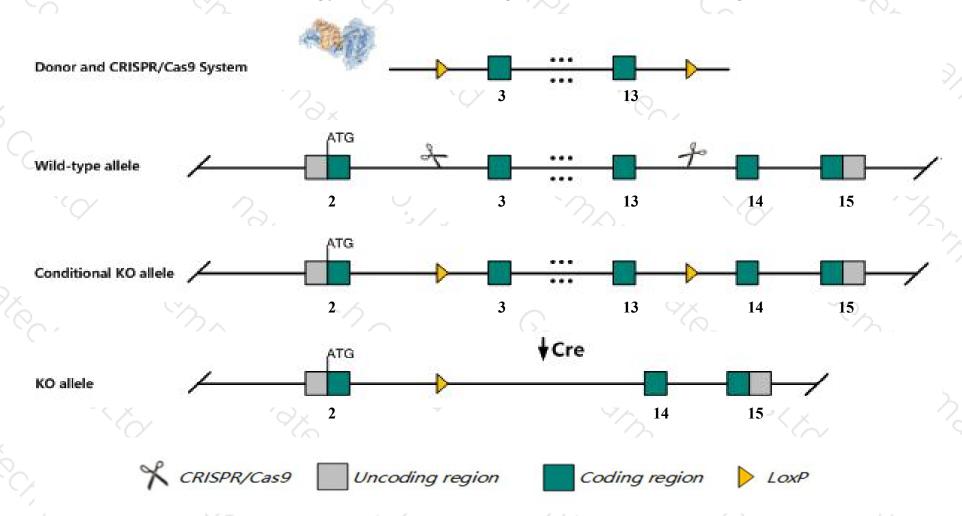
Strain background

C57BL/6JGpt

Conditional Knockout strategy



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



Technical routes



- The *Vcan* gene has 9 transcripts. According to the structure of *Vcan* gene, exon3-exon13 of *Vcan-203*(ENSMUST00000109546.8) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Vcan* 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, homozygotes for an insertional mutation exhibit anterior-posterior segmental defects of the heart, lack endocardial cushions of the conus and atrioventricular region, and die and around embryonic day 10.5.
- The *Vcan* gene is located on the Chr13. 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.
- ➤ The deleted area contains GM16318-201.
- ➤ The KO region deletes most of the coding sequence, but does not result in frameshift.

Gene information (NCBI)



Vcan versican [Mus musculus (house mouse)]

Gene ID: 13003, updated on 13-Mar-2020

Summary

↑ ?

Official Symbol Vcan provided by MGI

Official Full Name versican provided by MGI

Primary source MGI:MGI:102889

See related Ensembl: ENSMUSG00000021614

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 5430420N07Rik, 9430051N09, Cspq2, DPEAAE, NG2, PG-M, PG-M(V0), PG-M(V1), hdf

Expression Biased expression in limb E14.5 (RPKM 25.2), CNS E11.5 (RPKM 14.1) and 9 other tissuesSee more

Orthologs <u>human all</u>

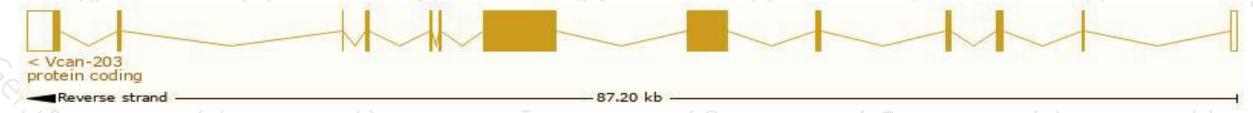
Transcript information (Ensembl)



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

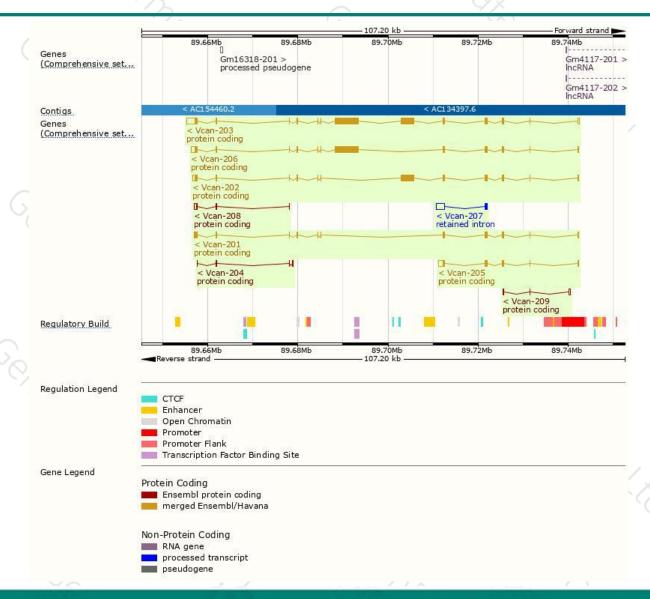
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Vcan-203	ENSMUST00000109546.8	12427	3354aa	Protein coding	CCDS36740	E9PYH0	TSL:1 GENCODE basic
Vcan-206	ENSMUST00000159910.7	8332	2394aa	Protein coding	CCDS49323	<u>G3XA35</u>	TSL:5 GENCODE basic
Vcan-202	ENSMUST00000109544.8	5702	1615aa	Protein coding	CCDS49324	E9QMK2	TSL:5 GENCODE basic
Vcan-201	ENSMUST00000109543.8	2321	<u>655aa</u>	Protein coding	CCDS49322	E9QMK3	TSL:1 GENCODE basic APPRIS P1
Vcan-205	ENSMUST00000159337.7	2273	<u>368aa</u>	Protein coding	CCDS49321	Q8BS97	TSL:1 GENCODE basic
Vcan-208	ENSMUST00000160740.7	861	<u>141aa</u>	Protein coding	528	<u>F7B603</u>	CDS 5' incomplete TSL:2
Vcan-209	ENSMUST00000162715.1	615	<u>100aa</u>	Protein coding		E0CZC0	CDS 3' incomplete TSL:3
Vcan-204	ENSMUST00000159285.1	294	<u>98aa</u>	Protein coding	020	F7B6F7	CDS 5' and 3' incomplete TSL:5
Vcan-207	ENSMUST00000160029.1	2144	No protein	Retained intron	(5)	-	TSL:1
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The strategy is based on the design of *Vcan-203* transcript, the transcription is shown below:



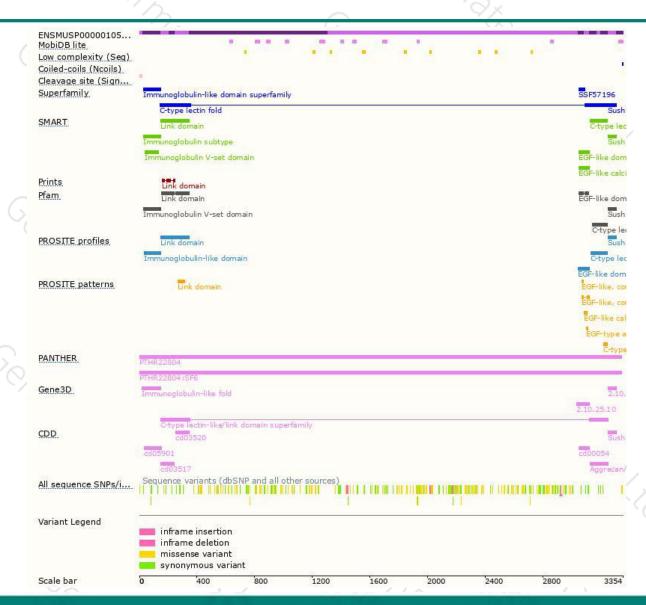
Genomic location distribution





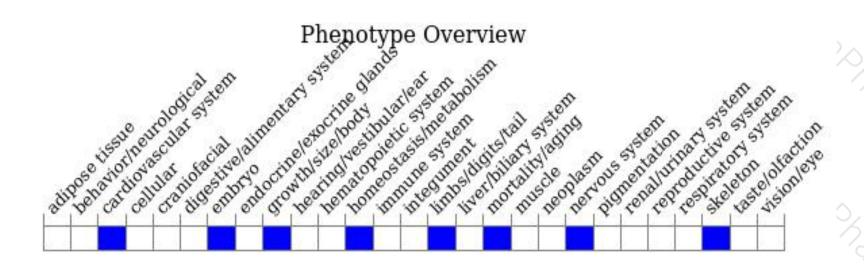
Protein domain





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, homozygotes for an insertional mutation exhibit anterior-posterior segmental defects of the heart, lack endocardial cushions of the conus and atrioventricular region, and die and around embryonic day 10.5.



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





