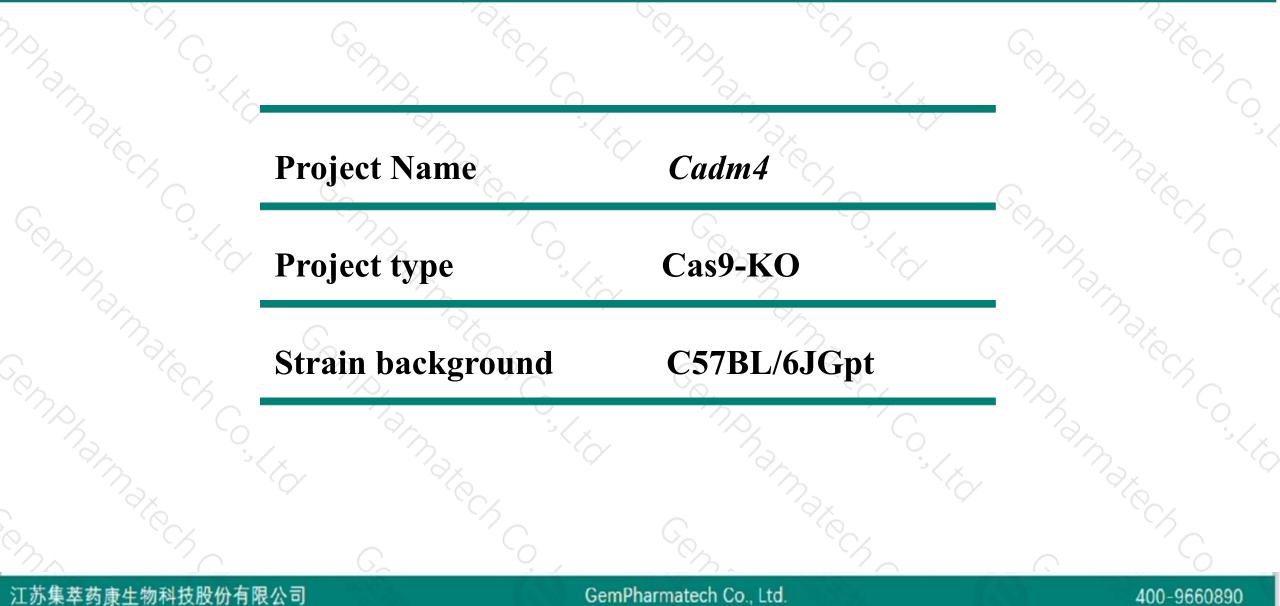


Cadm4 Cas9-KO Strategy

Designer: Xueting Zhang Reviewer: Yanhua Shen Date: 2020-02-24

Project Overview

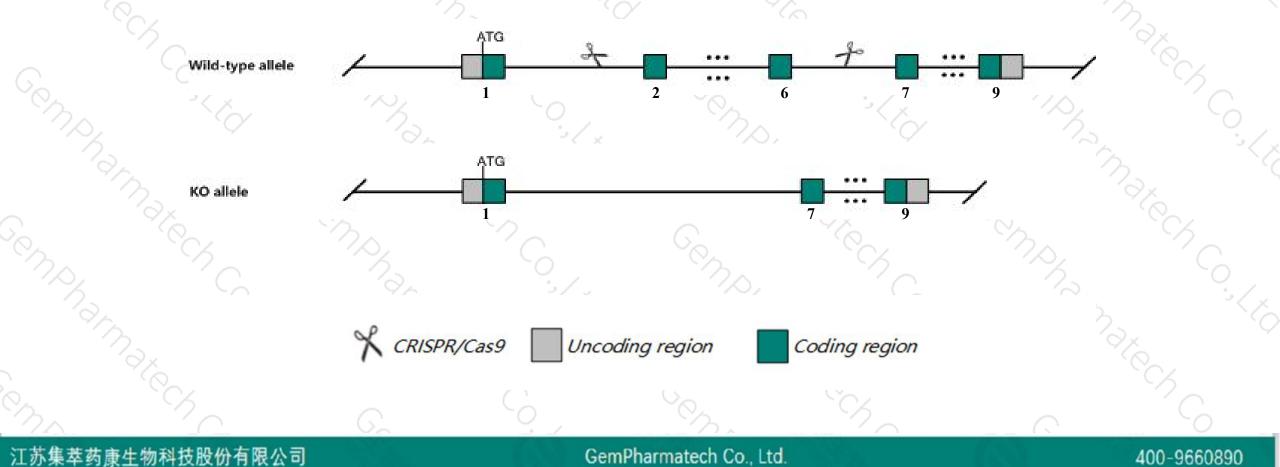




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Cadm4 gene. The schematic diagram is as follows:





- The Cadm4 gene has 2 transcripts. According to the structure of Cadm4 gene, exon2-exon6 of Cadm4-201 (ENSMUST0000068023.7) transcript is recommended as the knockout region. The region contains 691bp coding sequence. Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify Cadm4 gene. The brief process is as follows: CRISPR/Cas9 system

- According to the existing MGI data, Mice homozygous for one null allele do not display myelination abnormalities. Mice with ubiquitous conditional deletion of the gene show myelination abnormalities, decreased nerve conduction velocity, hindlimb rigidity, limb grasping, and impaired coordination.
- The knockout region is near to the N-terminal of Zfp428 gene, this strategy may influence the regulatory function of the N-terminal of Zfp428 gene.
- The Cadm4 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Notice

Gene information (NCBI)

Official Symbol Cadm4 provided by MGI



Maxe Cr

Cadm4 cell adhesion molecule 4 [Mus musculus (house mouse)]

Gene ID: 260299, updated on 21-Aug-2019

Summary

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Official Full Name	cell adhesion molecule 4 provided by MGI
Primary source	MGI:MGI:2449088
See related	Ensembl:ENSMUSG00000054793
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Tsll2; lgdf4c; lgsf4c
Expression	Biased expression in CNS E18 (RPKM 63.0), cerebellum adult (RPKM 62.2) and 14 other tissues See more
Orthologs	human all

Genomic context

2 ?

See Cadm4 in Genome Data Viewer

Exon count: 9

Location: 7; 7 A3

Annotation release	Status	Assembly	Chr	Location	24
108	current	GRCm38.p6 (GCF_000001635.26)	7	NC_000073.6 (2448202324504533)	1
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	7	NC_000073.5 (2526704225289552)	

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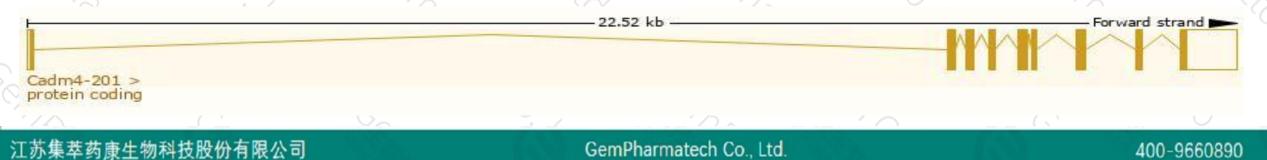
Transcript information (Ensembl)



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

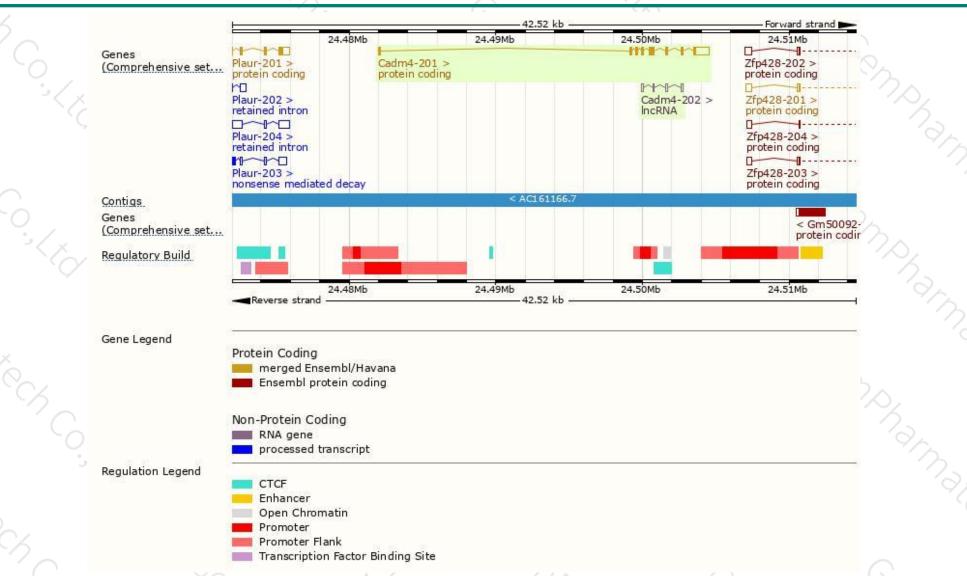
Name Transcript ID		bp	Protein	Biotype	CCDS	UniProt	Flags	
Cadm4-201	ENSMUST0000068023.7	2161	<u>388aa</u>	Protein coding	CCDS20951	<u>Q8R464</u>	TSL:1 GENCODE basic APPRIS P1	
Cadm4-202	ENSMUST00000205820.1	491	No protein	IncRNA	87		TSL:3	

The strategy is based on the design of Cadm4-201 transcript, The transcription is shown below



Genomic location distribution





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Protein domain



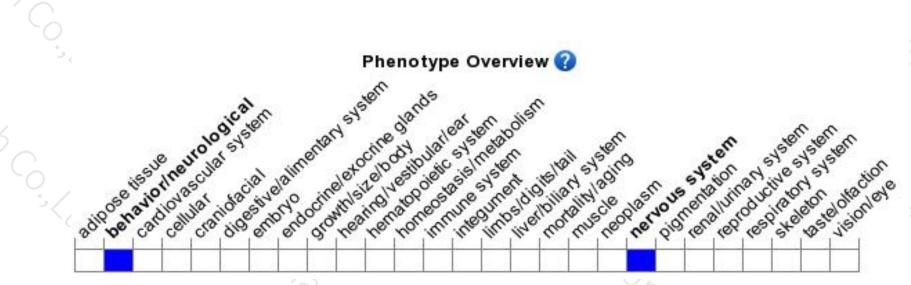
ENSMUSP00000066 Transmembrane heli SIFTS import Low complexity (Seg)								-	- 5	S.C.
Cleavage site (Sign Superfamily SMART		obulin-like domain	superfamily							3
Pfam		lobulin subtype Jobulin V-set doma	and the second se	nunoglobulin su -like, immunogl		at			Neurexin/synder	
	and the second	Depending of the bearing			P	¥13927				0.2
PROSITE profiles PANTHER	Cell adhesion m	volecule 4	Immuno	oglobulin-like de	omain					
Gene3D CDD	PTHR45889 Immunoglo	bulin-like fold	- it-							1
All sequence SNPs/i	Sequence varian	ts (dbSNP and al	l other sour	ces)		cd00096	r.	10) ''''()
Variant Legend	synonymou	s variant		160		-37	100			
Scale bar	o 40	80	120	160	200	240	280	320	388	
A 10		~ <u>0</u>	/	So.		2				

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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 one null allele do not display myelination abnormalities. Mice with ubiquitous conditional deletion of the gene show myelination abnormalities, decreased nerve conduction velocity, hindlimb rigidity, limb grasping, and impaired coordination.

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



