

Cdh23 Cas9-CKO Strategy

Designer: Huimin Su

Reviewer: Ruirui Zhang

Design Date: 2020/1/22

Project Overview



Project Name

Cdh23

Project type

Cas9-CKO

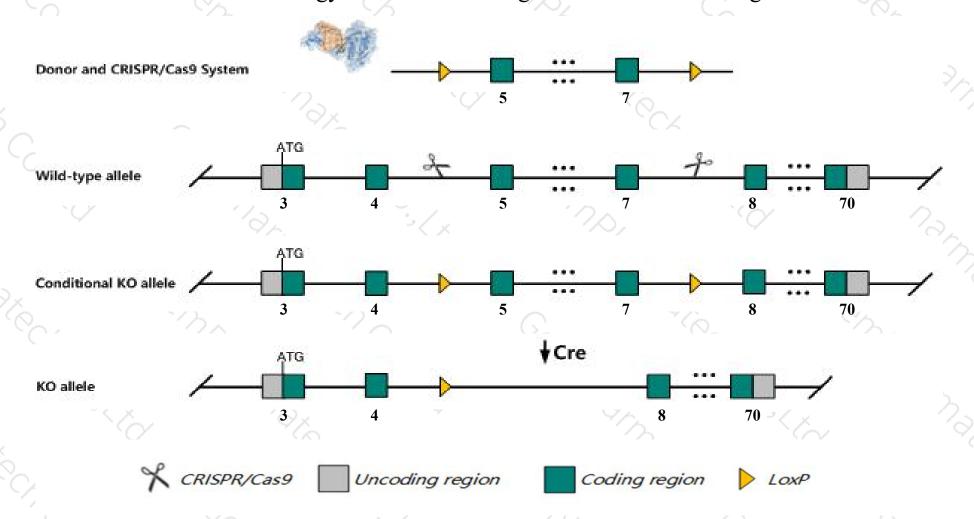
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Cdh23* gene. The schematic diagram is as follows:



Technical routes



- The *Cdh23* gene has 16 transcripts. According to the structure of *Cdh23* gene, exon5-exon7 of *Cdh23-205* (ENSMUST00000105464.8) transcript is recommended as the knockout region. The region contains 284bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cdh23* 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, Mutant mice exhibit circling behavior, tilting of the head and are deaf.

 Mice homozygous for a targeted knock-out exhibit abnormal outer hair cells morphology.
- The *Cdh23* gene is located on the Chr10. 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)



Cdh23 cadherin 23 (otocadherin) [Mus musculus (house mouse)]

Gene ID: 22295, updated on 19-Nov-2019

Summary

Official Symbol Cdh23 provided by MGI

Official Full Name cadherin 23 (otocadherin) provided by MGI

Primary source MGI:MGI:1890219

See related Ensembl:ENSMUSG00000012819

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 v; ahl; bob; bus; ahl1; mdfw; sals; USH1D; nmf112; nmf181; nmf252; 4930542A03Rik

Expression Biased expression in testis adult (RPKM 8.3), heart adult (RPKM 1.0) and 8 other tissues See more

Orthologs <u>human</u> all

Genomic context

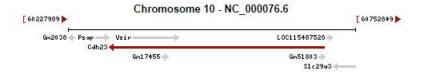
Location: 10 B4; 10 30.11 cM

See Cdh23 in Genome Data Viewer

△ ?

Exon count: 73

Annotation release	Status	Assembly	Chr	Location	
108	current	GRCm38.p6 (GCF_000001635.26)	10	NC_000076.6 (6030274860696513, complement)	
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	10	NC_000076.5 (5976602260159238, complement)	



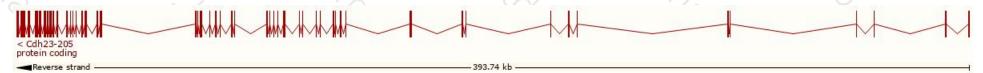
Transcript information (Ensembl)



The gene has 16 transcripts, all transcripts are shown below:

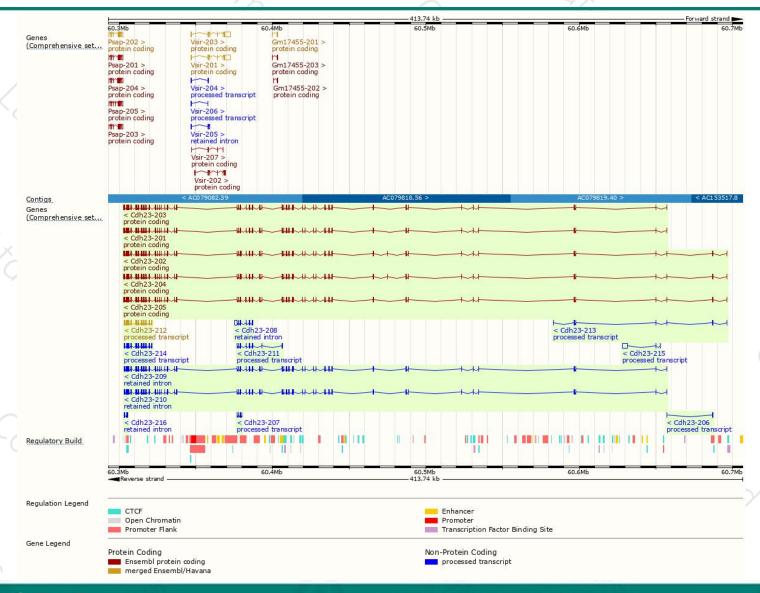
Name	Transcript ID 🍦	bp 👙	Protein 🍦	Biotype	CCDS 🍦	UniProt #		Flags	4	
Cdh23-205	ENSMUST00000105464.8	11090	<u>3352aa</u>	Protein coding	CCDS56704₽	<u>K4DI74</u> ₽	TSL:5	GENCODE basic	APPRIS P2	
Cdh23-202	ENSMUST00000105461.9	11096	3354aa	Protein coding	8	Q99PF4₽	TSL:5	GENCODE basic	APPRIS ALT2	
Cdh23-204	ENSMUST00000105463.8	10991	3319aa	Protein coding	8	Q99PF4₽	TSL:5	GENCODE basic	APPRIS ALT2	
Cdh23-201	ENSMUST00000073242.10	10688	3353aa	Protein coding	8	F8WIF5₽	TSL:5	GENCODE basic	APPRIS ALT2	
Cdh23-203	ENSMUST00000105462.7	10592	3321aa	Protein coding	8	E9Q7M6@	TSL:5	GENCODE basic	APPRIS ALT2	
Cdh23-212	ENSMUST00000151194.7	3787	No protein	Processed transcript	8	15	TSL:1			
Cdh23-214	ENSMUST00000153525.7	3682	No protein	Processed transcript	8	1.7	TSL:1			
Cdh23-215	ENSMUST00000153677.1	3041	No protein	Processed transcript	8	15	TSL:1			
Cdh23-211	ENSMUST00000144462.7	1510	No protein	Processed transcript	- 5	1.5	TSL:5			
Cdh23-213	ENSMUST00000153149.7	1052	No protein	Processed transcript		15	TSL:1			
Cdh23-207	ENSMUST00000124712.1	650	No protein	Processed transcript	- 50	1.7	TSL:3			
Cdh23-206	ENSMUST00000123309.1	438	No protein	Processed transcript		15	TSL:2			
Cdh23-210	ENSMUST00000135638.7	10435	No protein	Retained intron	8	1.5	TSL:5			
Cdh23-209	ENSMUST00000128249.7	10295	No protein	Retained intron	8	5	TSL:5			
Cdh23-208	ENSMUST00000127312.7	2574	No protein	Retained intron	- 5	15	TSL:5			
Cdh23-216	ENSMUST00000156501.1	1064	No protein	Retained intron	-	15		TSL:2		

The strategy is based on the design of Cdh23-205 transcript, The transcription is shown below



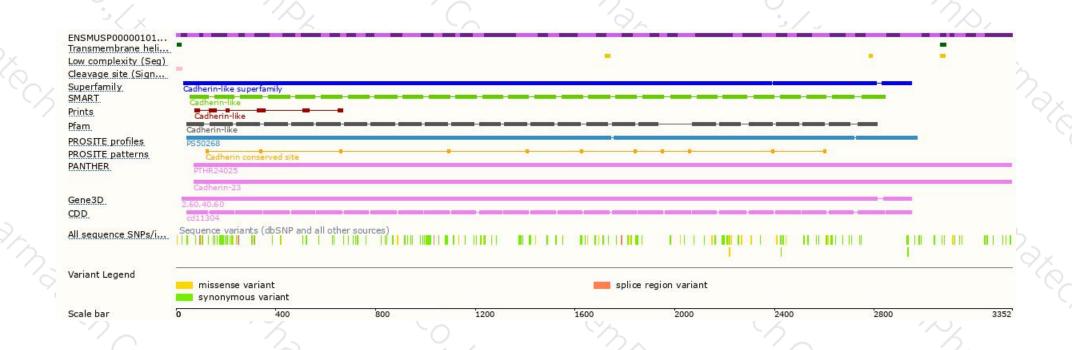
Genomic location distribution





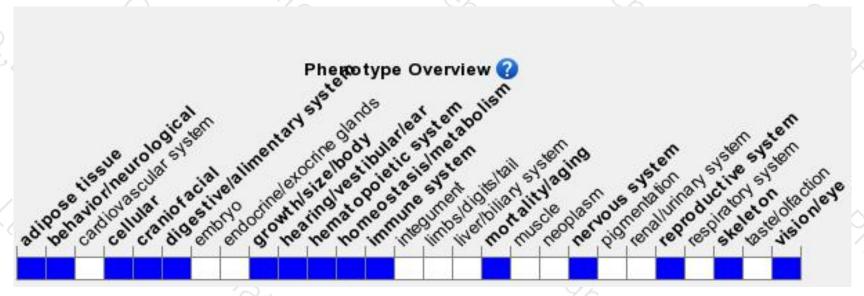
Protein domain





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, Mutant mice exhibit circling behavior, tilting of the head and are deaf. Mice homozygous for a targeted knock-out exhibit abnormal outer hair cells morphology.



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





