

Wdr74 Cas9-CKO Strategy

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Project Overview



Project Name

Wdr74

Project type

Cas9-CKO

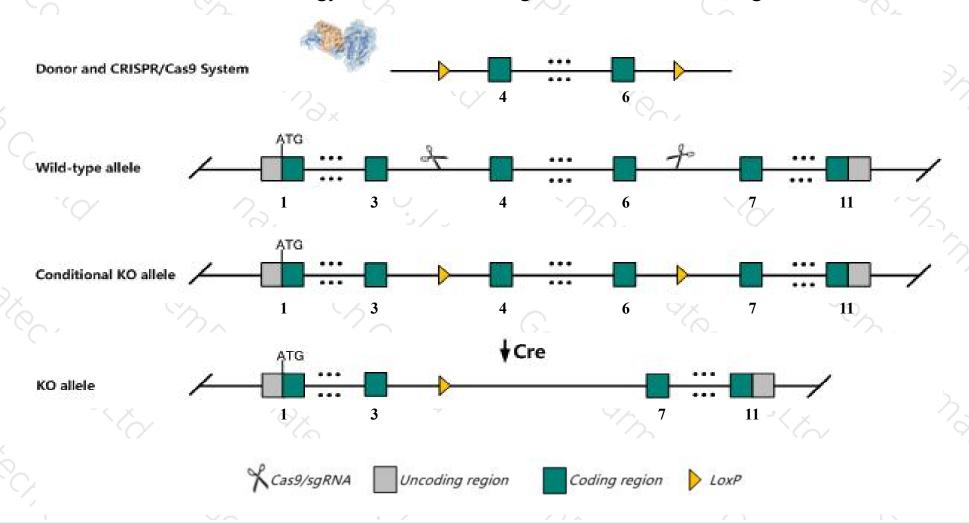
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Wdr74* gene. The schematic diagram is as follows:



Technical routes



- The *Wdr74* gene has 8 transcripts. According to the structure of *Wdr74* gene, exon4-exon6 of *Wdr74-201*(ENSMUST00000049424.10) transcript is recommended as the knockout region. The region contains 325bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Wdr74* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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



- \gt The *Wdr74* gene is located on the Chr19. 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.
- \rightarrow The N-terminal of Wdr74 gene will remain several amino acids ,it may remain the partial function of Wdr74 gene.
- > Transcript *Wdr74*-206&208 may not be affected.
- > The floxed region is near to the N-terminal of 1700092M07Rik gene and Stx5a gene Mir6992 gene, this strategy may influence the regulatory function of the N-terminal of these gene.
- 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)



Wdr74 WD repeat domain 74 [Mus musculus (house mouse)]

Gene ID: 107071, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Wdr74 provided by MGI

Official Full Name WD repeat domain 74 provided by MGI

Primary source MGI:MGI:2147427

See related Ensembl:ENSMUSG00000042729

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 5730436H21Rik, AA407588

Expression Ubiquitous expression in liver E14 (RPKM 17.4), liver E14.5 (RPKM 16.6) and 28 other tissuesSee more

Orthologs <u>human all</u>

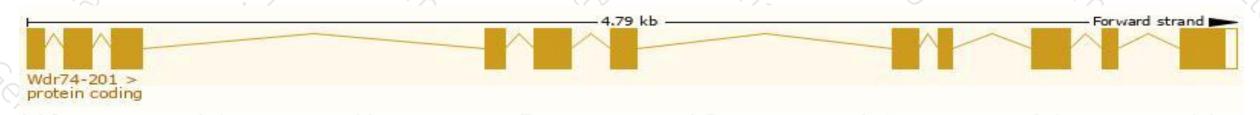
Transcript information (Ensembl)



The gene has 8 transcripts, all transcripts are shown below:

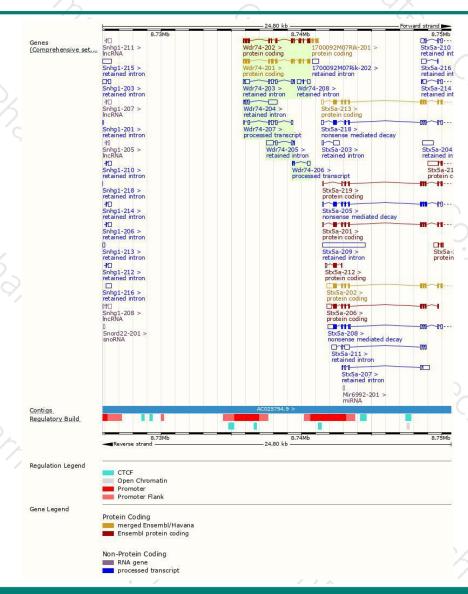
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Wdr74-201	ENSMUST00000049424.10	1204	384aa	Protein coding	CCDS29541	Q8VCG3	TSL:1 GENCODE basic APPRIS P1
Wdr74-202	ENSMUST00000210512.1	1078	<u>340aa</u>	Protein coding		Q3UL50	TSL:1 GENCODE basic
Wdr74-207	ENSMUST00000237673.1	572	No protein	Processed transcript	8 <u>2</u> 8	7/24	
Wdr74-206	ENSMUST00000237531.1	298	No protein	Processed transcript	-	(5)	
Wdr74-204	ENSMUST00000236946.1	855	No protein	Retained intron	-	140	
Wdr74-203	ENSMUST00000210592.2	846	No protein	Retained intron	878	15	TSL:5
Wdr74-205	ENSMUST00000237017.1	742	No protein	Retained intron	5÷	i e	
Wdr74-208	ENSMUST00000238116.1	577	No protein	Retained intron	12	-2	

The strategy is based on the design of *Wdr74-201* transcript, the transcription is shown below:



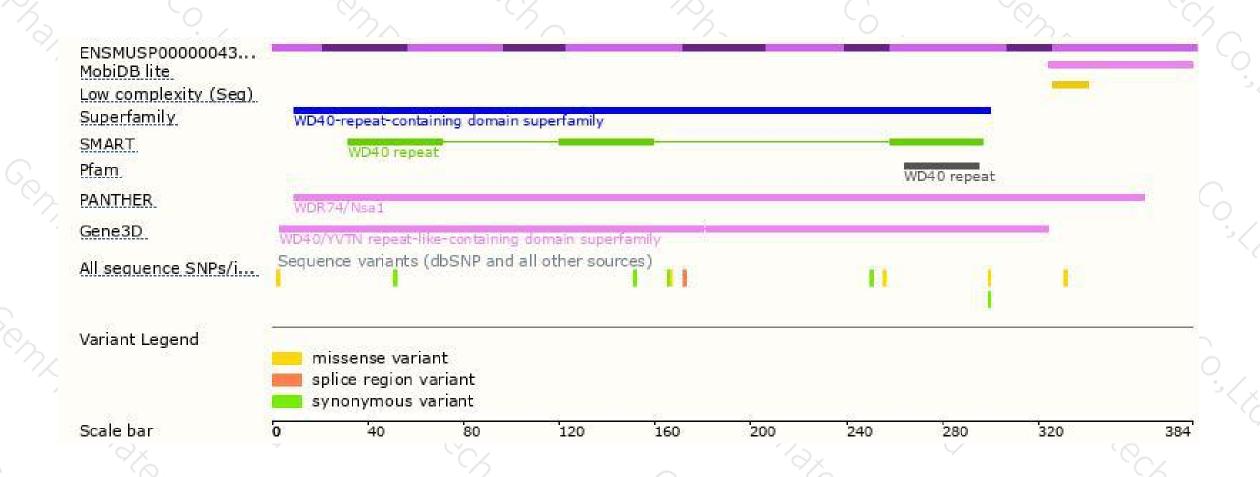
Genomic location distribution





Protein domain







If you have any questions, you are welcome to inquire.

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