

Ago1 Cas9-CKO Strategy

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Project Overview



Project Name

Ago1

Project type

Cas9-CKO

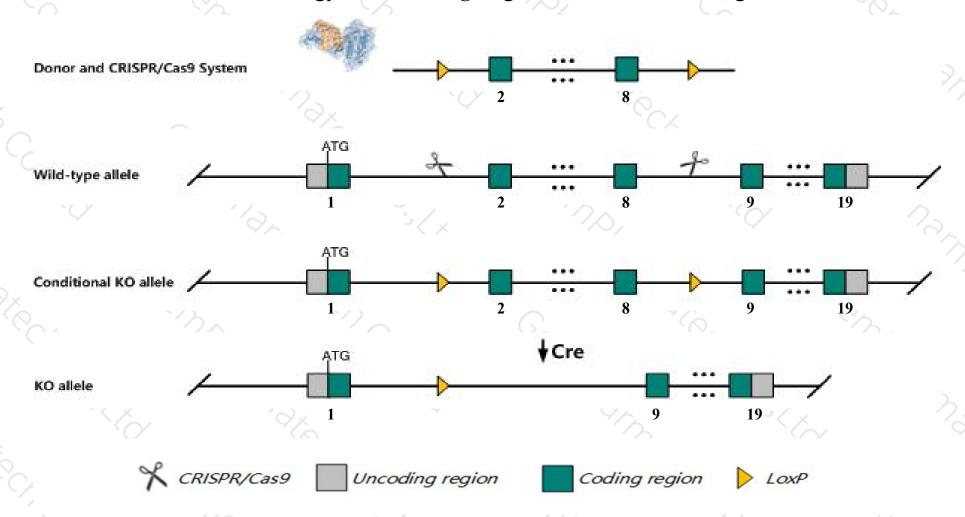
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Ago1 gene. The schematic diagram is as follows:



Technical routes



- ➤ The Ago1 gene has 5 transcripts. According to the structure of Ago1 gene, exon2-exon8 of Ago1-201

 (ENSMUST00000097888.9) transcript is recommended as the knockout region. The region contains 995bp coding sequence.

 Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Ago1* 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, Mice homozygous for a conditional allele activated in keratinocytes exhibit no abnormal phenotype.
- The *Ago1* gene is located on the Chr4. 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)



Ago1 argonaute RISC catalytic subunit 1 [Mus musculus (house mouse)]

Gene ID: 236511, updated on 31-Jan-2019

Summary

☆ ?

Official Symbol Ago1 provided by MGI

Official Full Name argonaute RISC catalytic subunit 1 provided by MGI

Primary source MGI:MGI:2446630

See related Ensembl: ENSMUSG00000041530

Gene type protein coding
RefSeq status REVIEWED

Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as Eif2c1

Summary This gene encodes a member of the argonaute family of proteins, which associate with small RNAs and have important roles in RNA

interference (RNAi) and RNA silencing. This protein binds to microRNAs (miRNAs) or small interfering RNAs (siRNAs) and represses translation of mRNAs that are complementary to them. It is also involved in transcriptional gene silencing (TGS) of promoter regions that are complementary to bound short antigene RNAs (agRNAs), as well as in the degradation of miRNA-bound mRNA targets. Alternatively spliced transcript variants encoding different isoforms have been found for this gene. A recent study showed this gene to be an authentic stop codon readthrough target, and that its mRNA could give rise to an additional C-terminally extended isoform by use of an alternative in-frame

translation termination codon. [provided by RefSeq, Nov 2015]

Expression Ubiquitous expression in CNS E11.5 (RPKM 22.9), CNS E14 (RPKM 19.3) and 28 other tissuesSee more

Orthologs <u>human</u> all

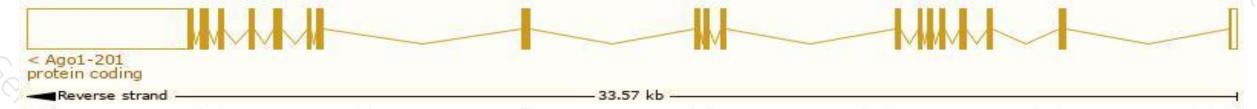
Transcript information (Ensembl)



The gene has 5 transcripts, all transcripts are shown below:

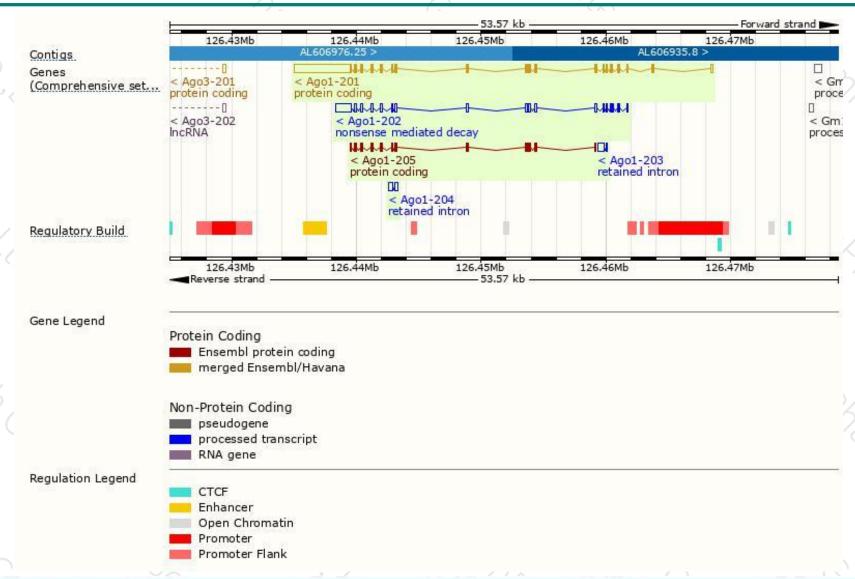
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ago1-201	ENSMUST00000097888.9	7227	857aa	Protein coding	CCDS18653	Q8CJG1	TSL:1 GENCODE basic APPRIS P1
Ago1-205	ENSMUST00000176315.1	1662	<u>553aa</u>	Protein coding	-	H3BJ70	CDS 5' incomplete TSL:5
Ago1-202	ENSMUST00000127800.1	3606	<u>133aa</u>	Nonsense mediated decay	28	H3BL59	CDS 5' incomplete TSL:1
Ago1-203	ENSMUST00000149425.1	678	No protein	Retained intron	29	15-25	TSL:3
Ago1-204	ENSMUST00000156533.1	588	No protein	Retained intron	-	1377	TSL:3

The strategy is based on the design of Ago1-201 transcript, The transcription is shown below



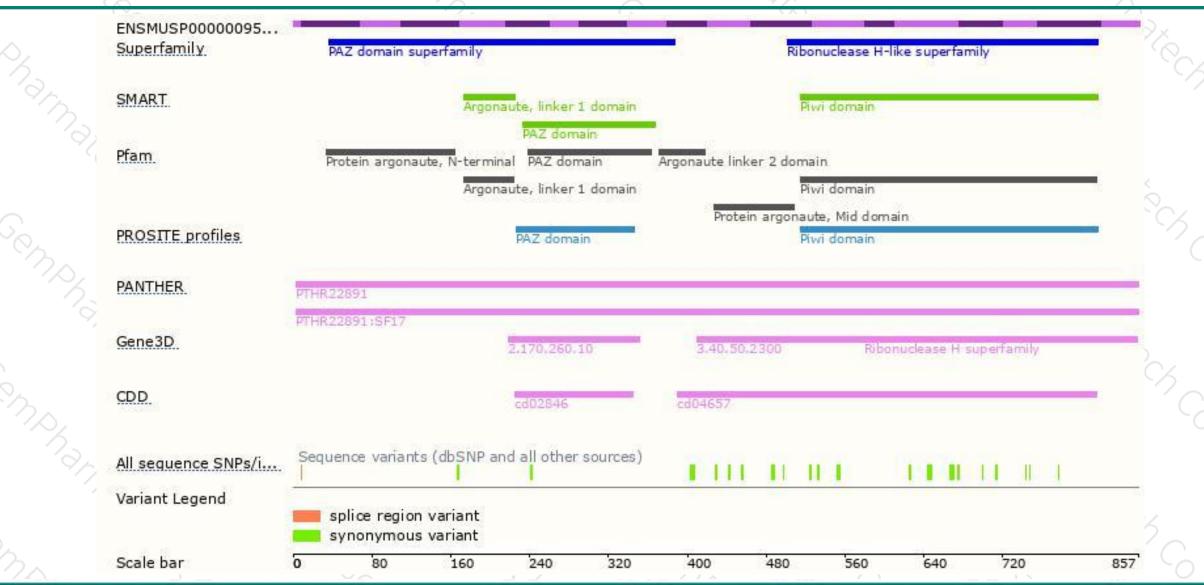
Genomic location distribution





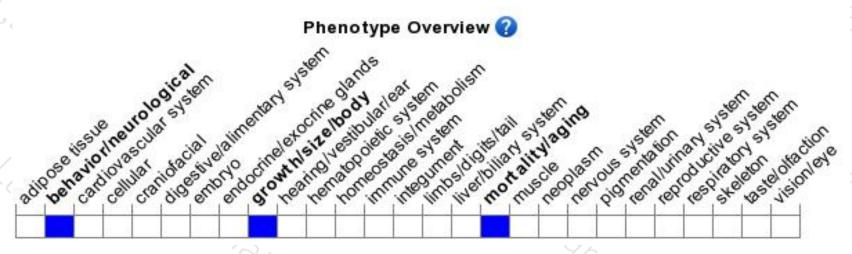
Protein domain





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, Mice homozygous for a conditional allele activated in keratinocytes exhibit no abnormal phenotype.



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





