

Arid1a Cas9-CKO Strategy

Designer:

Jinling Wang

Reviewer:

Lingyan Wu

Design Date:

2019-1-23

Project Overview



Project Name

Arid1a

Project type

Cas9-CKO

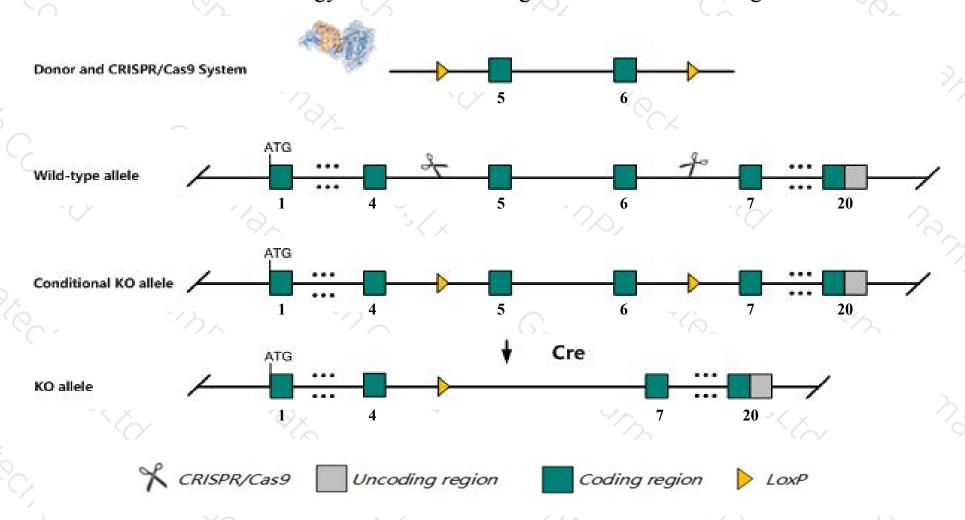
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Arid1a* gene. The schematic diagram is as follows:



Technical routes



- The *Arid1a* gene has 6 transcripts. According to the structure of *Arid1a* gene, exon5-exon6 of *Arid1a-202* (ENSMUST00000105897.9) transcript is recommended as the knockout region. The region contains 331bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Arid1a* 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, Embryos with homozygous null alleles arrest development at E6.5 with failure to form a mesoderm layer. Mice homozygous for an allele lacking exon 2 and 3 die shortly after birth and exhibit an increase in the hematopoietic stem cell population of the fetal liver at E14.5.
- > 641 amino acids will remain at the N-terminus and some functions may be retained.
- ➤ Transcript 204 is unaffected.
- The *Arid1a* 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)



Arid1a AT rich interactive domain 1A (SWI-like) [Mus musculus (house mouse)]

Gene ID: 93760, updated on 2-Apr-2019

Summary

☆ ?

Official Symbol Arid1a provided by MGI

Official Full Name AT rich interactive domain 1A (SWI-like) provided by MGI

Primary source MGI:MGI:1935147

See related Ensembl:ENSMUSG00000007880

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 1110030E03Rik, BAF250a, Osa1, Smarcf1

Expression Ubiquitous expression in thymus adult (RPKM 94.0), spleen adult (RPKM 52.9) and 28 other tissuesSee more

Orthologs <u>human</u> all

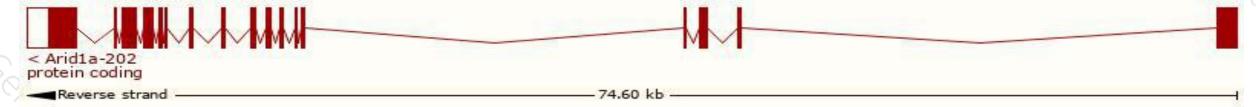
Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

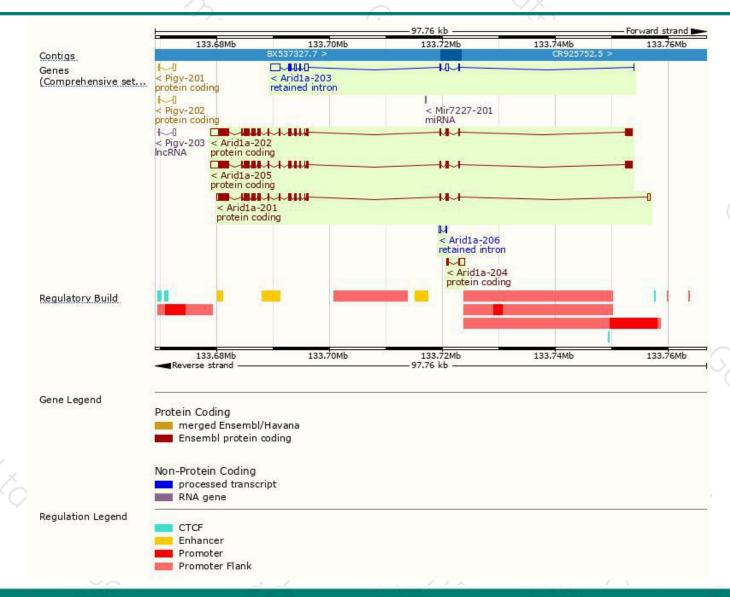
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Arid1a-202	ENSMUST00000105897.9	8175	2283aa	Protein coding	CCDS38908	A2BH40	TSL:5 GENCODE basic APPRIS P2
Arid1a-205	ENSMUST00000145664.8	8187	2287aa	Protein coding		E9QAQ7	TSL:2 GENCODE basic APPRIS ALT2
Arid1a-201	ENSMUST00000008024.6	6486	<u>1902aa</u>	Protein coding	120	A2BH40	TSL:1 GENCODE basic APPRIS ALT2
Arid1a-204	ENSMUST00000139709.2	1262	<u>140aa</u>	Protein coding	7527	E9Q495	CDS 3' incomplete TSL:2
Arid1a-203	ENSMUST00000138473.2	3964	No protein	Retained intron			TSL:1
Arid1a-206	ENSMUST00000168582.1	521	No protein	Retained intron	95 4 .3	-	TSL:2

The strategy is based on the design of Arid1a-202 transcript, The transcription is shown below



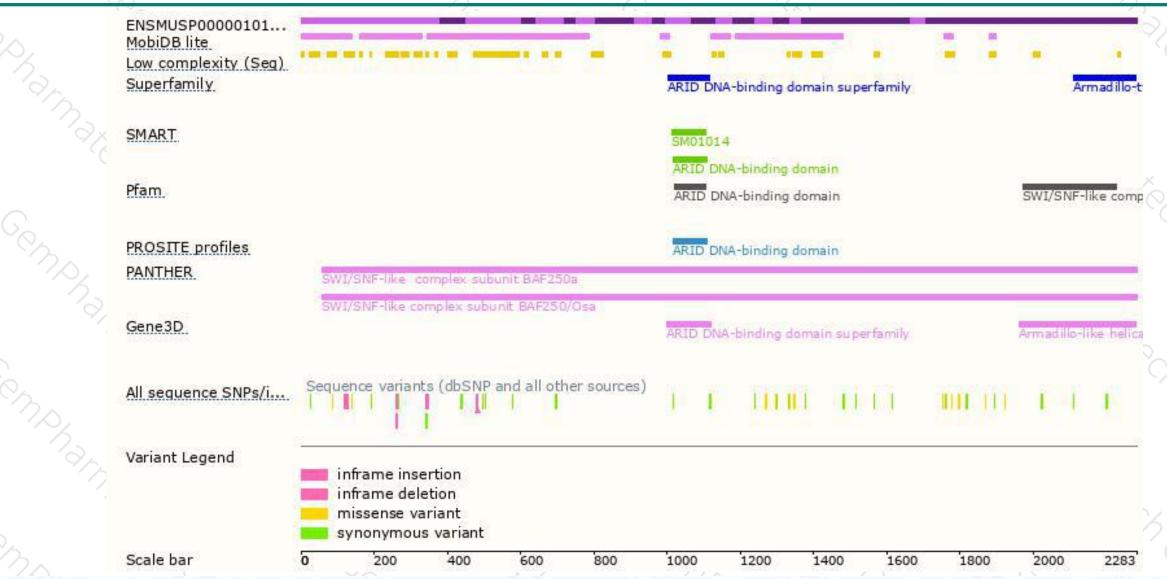
Genomic location distribution





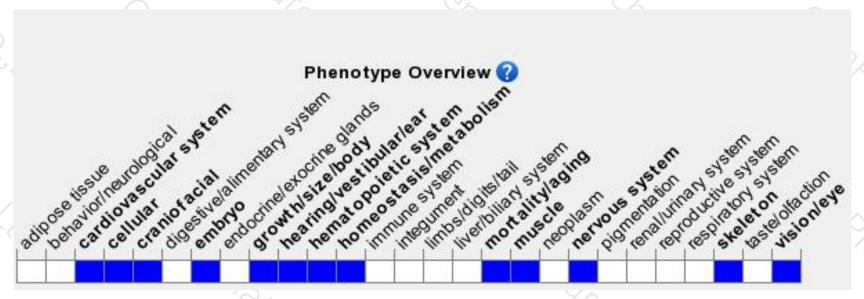
Protein domain





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, Embryos with homozygous null alleles arrest development at E6.5 with failure to form a mesoderm layer. Mice homozygous for an allele lacking exon 2 and 3 die shortly after birth and exhibit an increase in the hematopoietic stem cell population of the fetal liver at E14.5.



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





