Arid4b Cas9-CKO Strategy

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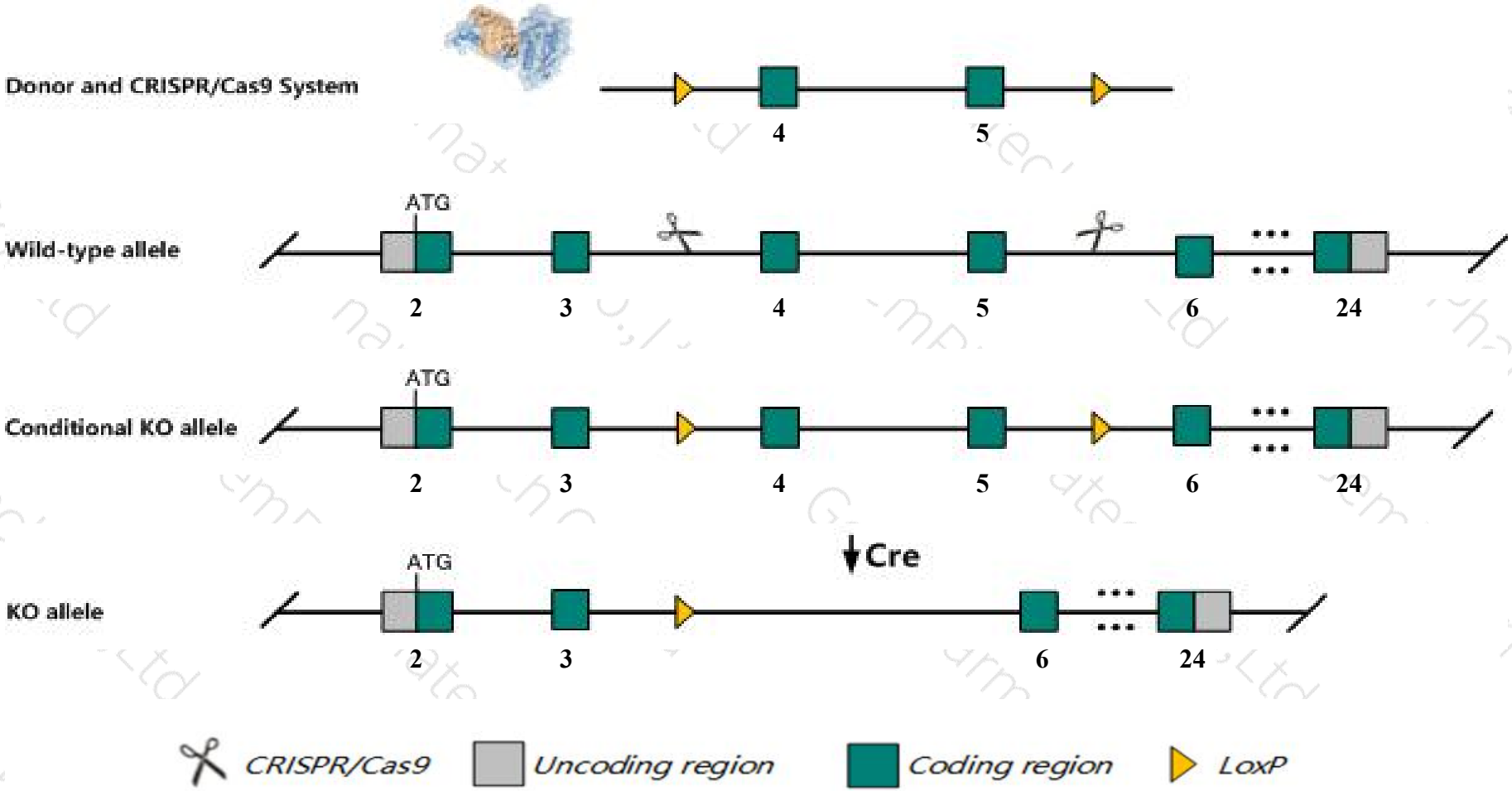
Design Date: 2020-7-24

Project Overview

Project Name	***Arid4b***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Arid4b* gene. The schematic diagram is as follows:



Technical routes

➢ The *Arid4b* gene has 12 transcripts. According to the structure of *Arid4b* gene, exon4-exon5 of *Arid4b-203*(ENSMUST00000110534.7) transcript is recommended as the knockout region.The region contains 157bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Arid4b* 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 null allele die pre-implantation.

➢ The *Arid4b* gene is located on the Chr13. 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）

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Arid4b AT rich interactive domain 4B (RBP1-like) [*Mus musculus* (house mouse)]

Gene ID: 94246, updated on 26-Jun-2020

Summary

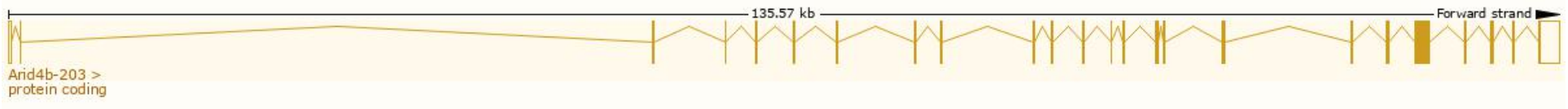
Official Symbol	Arid4b provided by MGI
Official Full Name	AT rich interactive domain 4B (RBP1-like) provided by MGI
Primary source	MGI:MGI:2137512
See related	Ensembl:ENSMUSG00000039219
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BCAA; BRCAA1; Rbp1l1; SAP180; RBBP1L1
Expression	Ubiquitous expression in CNS E11.5 (RPKM 10.1), CNS E14 (RPKM 7.3) and 26 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

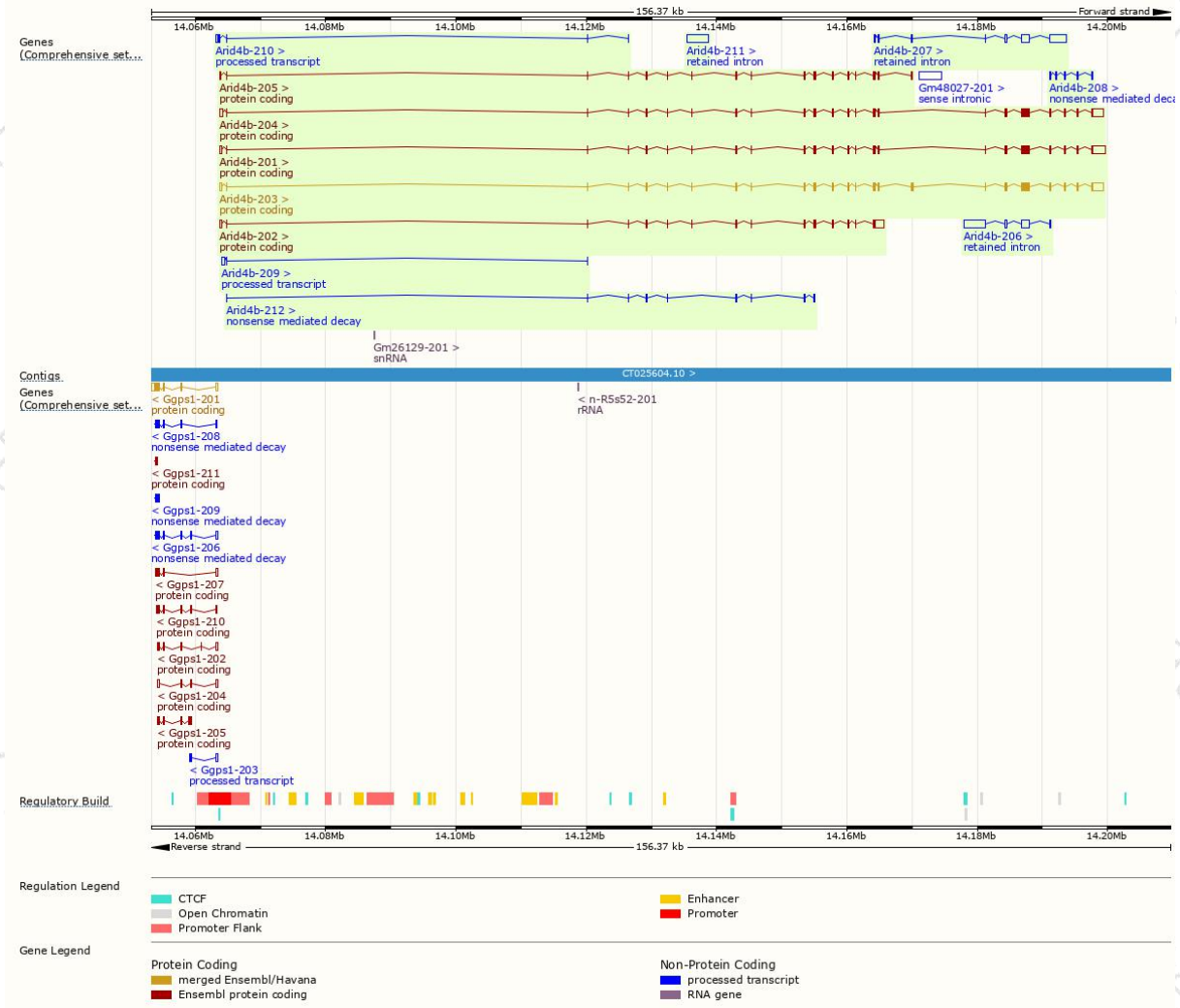
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein ▲	Biotype	CCDS	UniProt	Flags
Arid4b-208	ENSMUST00000151151.1	745	31aa	Nonsense mediated decay	-	A0A1Y7VKA9	CDS 5' incomplete TSL:3
Arid4b-212	ENSMUST00000222928.1	815	128aa	Nonsense mediated decay	-	A0A1Y7VIX3	TSL:5
Arid4b-202	ENSMUST00000110533.1	3020	473aa	Protein coding	-	Z4YL44	TSL:1 GENCODE basic
Arid4b-205	ENSMUST00000129488.7	1802	540aa	Protein coding	-	Z4YMH1	CDS 3' incomplete TSL:1
Arid4b-201	ENSMUST00000039538.14	5844	1227aa	Protein coding	CCDS36600	A2CG63	TSL:5 GENCODE basic
Arid4b-204	ENSMUST00000110536.7	5783	1227aa	Protein coding	CCDS36600	A2CG63	TSL:5 GENCODE basic
Arid4b-203	ENSMUST00000110534.7	5864	1314aa	Protein coding	CCDS36599	A2CG63	TSL:1 GENCODE basic APPRIS P1
Arid4b-209	ENSMUST00000151182.1	678	No protein	Processed transcript	-	-	TSL:3
Arid4b-210	ENSMUST00000155553.7	571	No protein	Processed transcript	-	-	TSL:3
Arid4b-206	ENSMUST00000148941.1	4828	No protein	Retained intron	-	-	TSL:2
Arid4b-207	ENSMUST00000149579.2	4571	No protein	Retained intron	-	-	TSL:5
Arid4b-211	ENSMUST00000222346.1	3353	No protein	Retained intron	-	-	TSL:NA

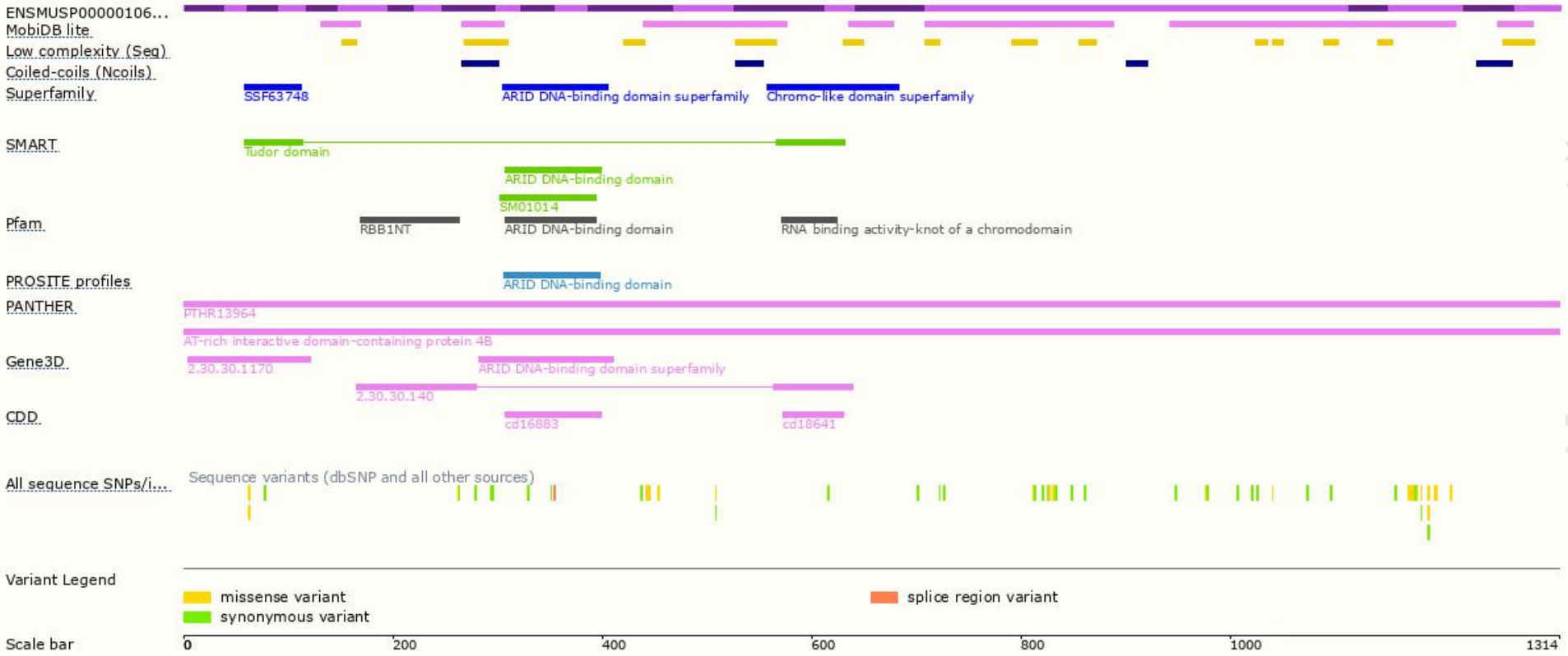
The strategy is based on the design of *Arid4b-203* transcript,the transcription is shown below:



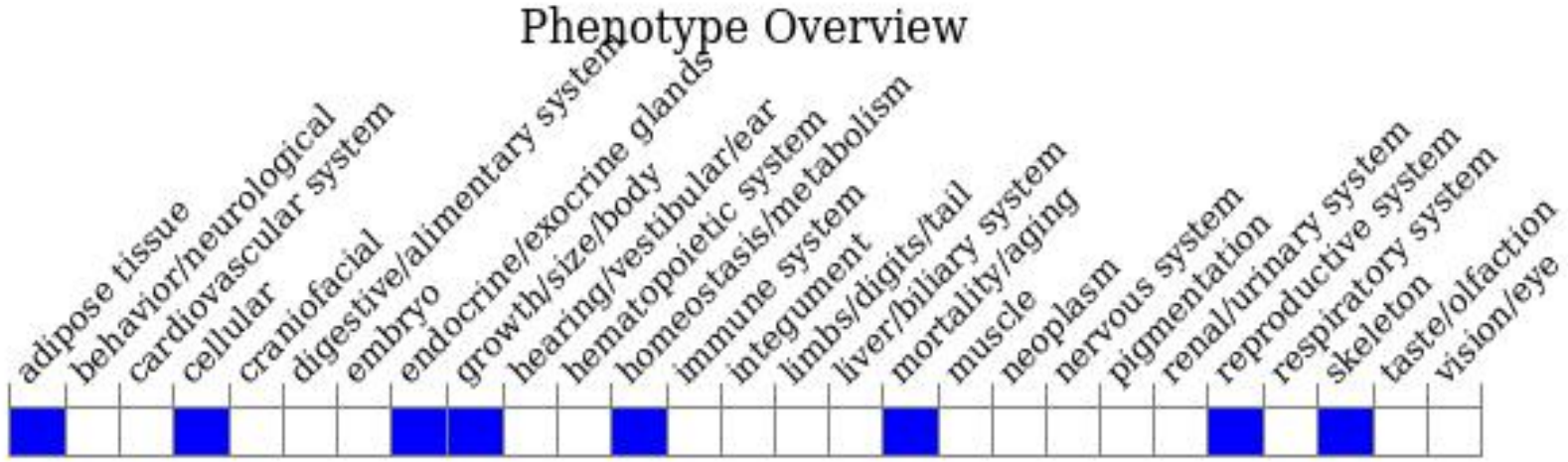
Genomic location distribution



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 null allele die pre-implantation.

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

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