

# Arhgef12 Cas9-CKO Strategy

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Reviewer: Daohua Xu

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# **Project Overview**



**Project Name** 

Arhgef12

**Project type** 

Cas9-CKO

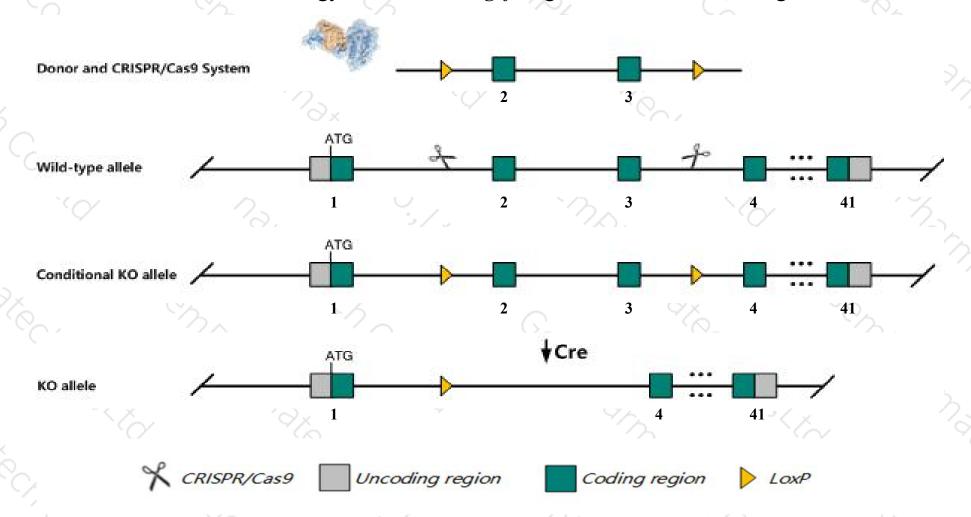
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Arhgef12 gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Arhgef12* gene has 6 transcripts. According to the structure of *Arhgef12* gene, exon2-exon3 of *Arhgef12-202*(ENSMUST00000165665.8) transcript is recommended as the knockout region. The region contains 110bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Arhgef12* 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 exhibit decreased sensitivity to certain vasoconstrictors and resistance to salt-induced hypertension. Mice homozygous for a different knock-out allele exhibit partial prenatal lethality.
- > Transcript *Arhgef12*-202&206 may not be affected.
- > The Arhgef12 gene is located on the Chr9. 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)



Arhgef12 Rho guanine nucleotide exchange factor (GEF) 12 [Mus musculus (house mouse)]

Gene ID: 69632, updated on 13-Mar-2020





Official Symbol Arhaef 12 provided by MGI

Official Full Name Rho quanine nucleotide exchange factor (GEF) 12 provided by MGI

Primary source MGI:MGI:1916882

See related Ensembl: ENSMUSG000000059495

Gene type protein coding

RefSeq status VALIDATED

Organism <u>Mus musculus</u>

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

Muroidea; Muridae; Murinae; Mus; Mus

Also known as 2310014B11Rik, AU019857, Larg, mKIAA0382

Expression Ubiquitous expression in lung adult (RPKM 23.6), kidney adult (RPKM 17.6) and 28 other tissuesSee more

Orthologs <u>human</u> <u>all</u>

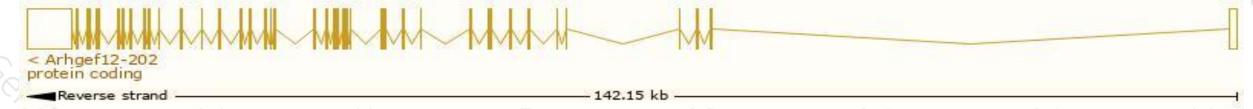
# Transcript information (Ensembl)



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

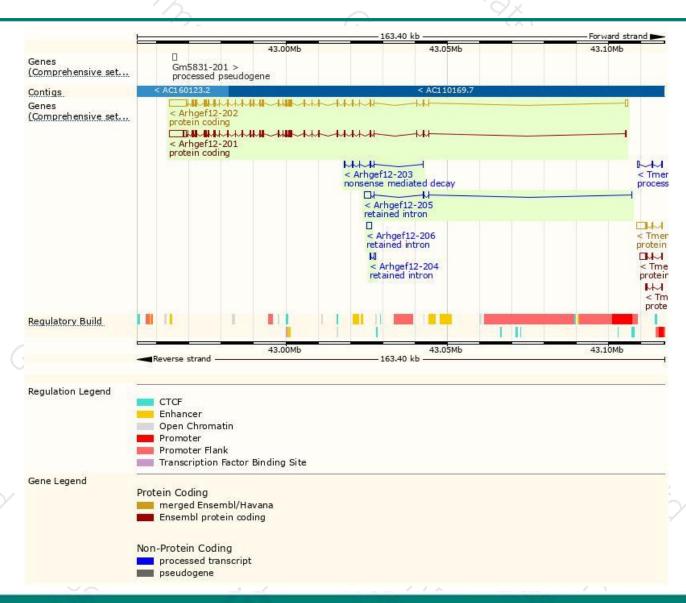
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Arhgef12-202	ENSMUST00000165665.8	10752	1544aa	Protein coding	CCDS40595	F8VQN6	TSL:1 GENCODE basic APPRIS P2
Arhgef12-201	ENSMUST00000072767.6	10059	1543aa	Protein coding	9 <del>-</del>	Q8R4H2	TSL:1 GENCODE basic APPRIS ALT1
Arhgef12-203	ENSMUST00000213566.1	555	<u>106aa</u>	Nonsense mediated decay	828	A0A1L1SUP5	CDS 5' incomplete TSL:3
Arhgef12-205	ENSMUST00000215898.1	2169	No protein	Retained intron	-	A	TSL:1
Arhgef12-206	ENSMUST00000217502.1	1714	No protein	Retained intron	-	ч	TSL:NA
Arhgef12-204	ENSMUST00000214175.1	848	No protein	Retained intron	1978	8	TSL:3

The strategy is based on the design of *Arhgef12-202* transcript, the transcription is shown below:



## Genomic location distribution





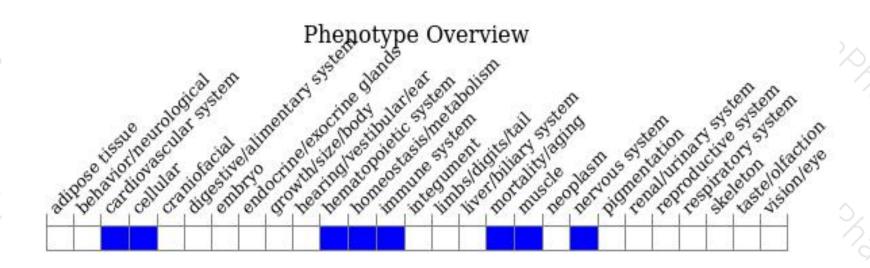
### Protein domain





## 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,mice homozygous for a null allele exhibit decreased sensitivity to certain vasoconstrictors and resistance to salt-induced hypertension. Mice homozygous for a different knock-out allele exhibit partial prenatal lethality.



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





