

Arf4 Cas9-KO Strategy

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Design Date: 2020-7-17

Project Overview



Project Name

Arf4

Project type

Cas9-KO

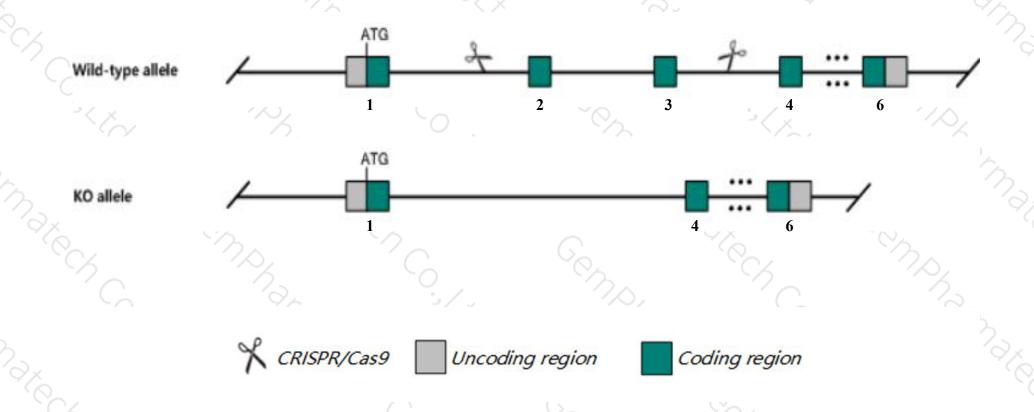
Strain background

C57BL/6JGpt

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Arf4 gene. The schematic diagram is as follows:



Technical routes



- > The Arf4 gene has 7 transcripts. According to the structure of Arf4 gene, exon2-exon3 of Arf4201(ENSMUST00000022429.8) transcript is recommended as the knockout region. The region contains 191bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Arf4* gene. The brief process is as follows: CRISPR/Cas9 system 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.

Notice



- > According to the existing MGI data, mice homozygous for a gene trap allele exhibit embryonic lethality. Mice heterozygous for the gene trap allele exhibit failure to increase exploration of an object in a novel context, reduced spine density and reduced mESPC amplitudes.
- > The Arf4 gene is located on the Chr14. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Arf4 ADP-ribosylation factor 4 [Mus musculus (house mouse)]

Gene ID: 11843, updated on 26-Mar-2020

Summary

☆ ?

Official Symbol Arf4 provided by MGI

Official Full Name ADP-ribosylation factor 4 provided by MGI

Primary source MGI:MGI:99433

See related Ensembl:ENSMUSG00000021877

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 AA407803

Expression Ubiquitous expression in adrenal adult (RPKM 186.8), duodenum adult (RPKM 168.4) and 28 other tissuesSee more

Orthologs human all

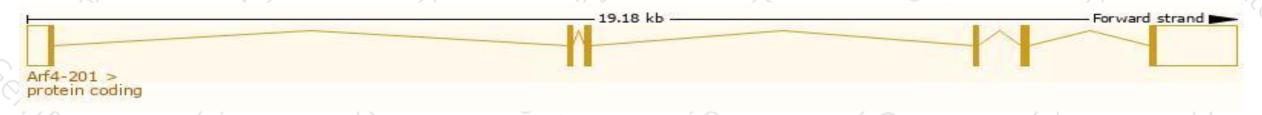
Transcript information (Ensembl)



The gene has 7 transcripts, all transcripts are shown below:

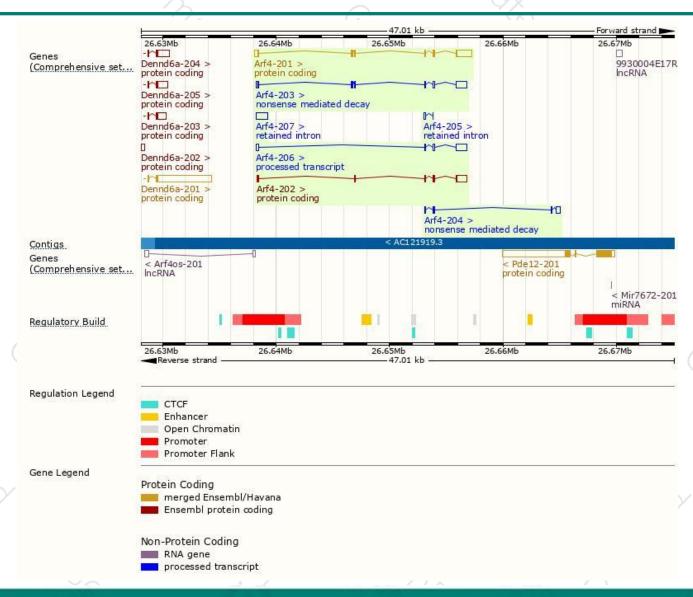
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Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST00000022429.8	2165	<u>180aa</u>	Protein coding	CCDS26881	P61750 Q14BR4	TSL:1 GENCODE basic APPRIS P1
ENSMUST00000112318.9	1367	<u>153aa</u>	Protein coding	0-0	E9Q798	TSL:5 GENCODE basic
ENSMUST00000166075.7	1582	<u>64aa</u>	Nonsense mediated decay	-	E9Q2C2	TSL:5
ENSMUST00000167376.1	700	<u>60aa</u>	Nonsense mediated decay	123	F6UFB9	CDS 5' incomplete TSL:5
ENSMUST00000171282.1	1345	No protein	Processed transcript	-	-	TSL:5
ENSMUST00000224138.1	1034	No protein	Retained intron	-	-	
ENSMUST00000171207.1	188	No protein	Retained intron		=	TSL:5
	ENSMUST00000112318.9 ENSMUST00000166075.7 ENSMUST00000167376.1 ENSMUST00000171282.1 ENSMUST00000224138.1	ENSMUST00000022429.8 2165 ENSMUST00000112318.9 1367 ENSMUST00000166075.7 1582 ENSMUST00000167376.1 700 ENSMUST00000171282.1 1345 ENSMUST00000224138.1 1034	ENSMUST00000022429.8 2165 180aa ENSMUST00000112318.9 1367 153aa ENSMUST00000166075.7 1582 64aa ENSMUST00000167376.1 700 60aa ENSMUST00000171282.1 1345 No protein ENSMUST000000224138.1 1034 No protein	ENSMUST00000022429.8 2165 180aa Protein coding ENSMUST00000112318.9 1367 153aa Protein coding ENSMUST00000166075.7 1582 64aa Nonsense mediated decay ENSMUST00000167376.1 700 60aa Nonsense mediated decay ENSMUST00000171282.1 1345 No protein Processed transcript ENSMUST000000224138.1 1034 No protein Retained intron	ENSMUST00000022429.8 2165 180aa Protein coding CCDS26881 ENSMUST00000112318.9 1367 153aa Protein coding - ENSMUST00000166075.7 1582 64aa Nonsense mediated decay - ENSMUST00000167376.1 700 60aa Nonsense mediated decay - ENSMUST00000171282.1 1345 No protein Processed transcript - ENSMUST000000224138.1 1034 No protein Retained intron -	ENSMUST00000022429.8 2165 180aa Protein coding CCDS26881 P61750 Q14BR4 ENSMUST00000112318.9 1367 153aa Protein coding - E9Q798 ENSMUST00000166075.7 1582 64aa Nonsense mediated decay - E9Q2C2 ENSMUST00000167376.1 700 60aa Nonsense mediated decay - F6UFB9 ENSMUST00000171282.1 1345 No protein Processed transcript - - ENSMUST00000224138.1 1034 No protein Retained intron - -

The strategy is based on the design of Arf4-201 transcript, the transcription is shown below:



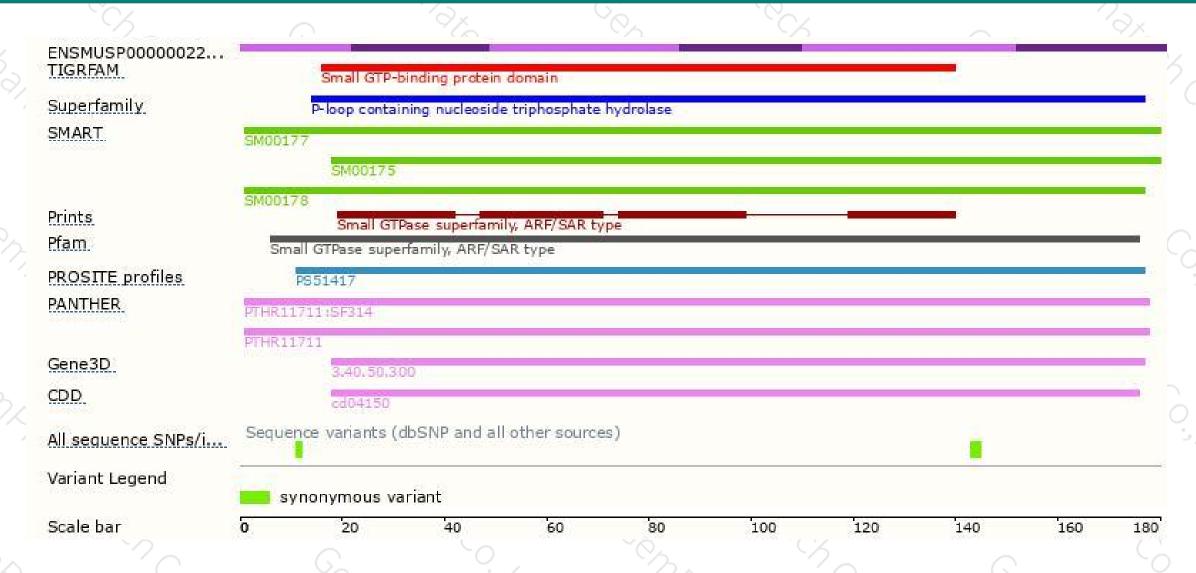
Genomic location distribution





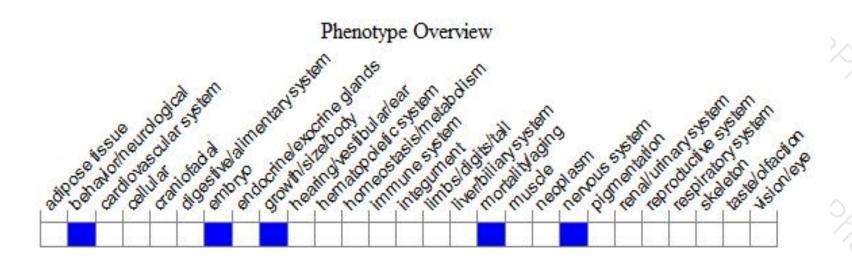
Protein domain





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 gene trap allele exhibit embryonic lethality. Mice heterozygous for the gene trap allele exhibit failure to increase exploration of an object in a novel context, reduced spine density and reduced mESPC amplitudes.



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





