

Acp5 Cas9-CKO Strategy

Designer: Huan Wang

Reviewer: Huan Fan

Design Date: 2020-5-6

Project Overview



Project Name

Acp5

Project type

Cas9-CKO

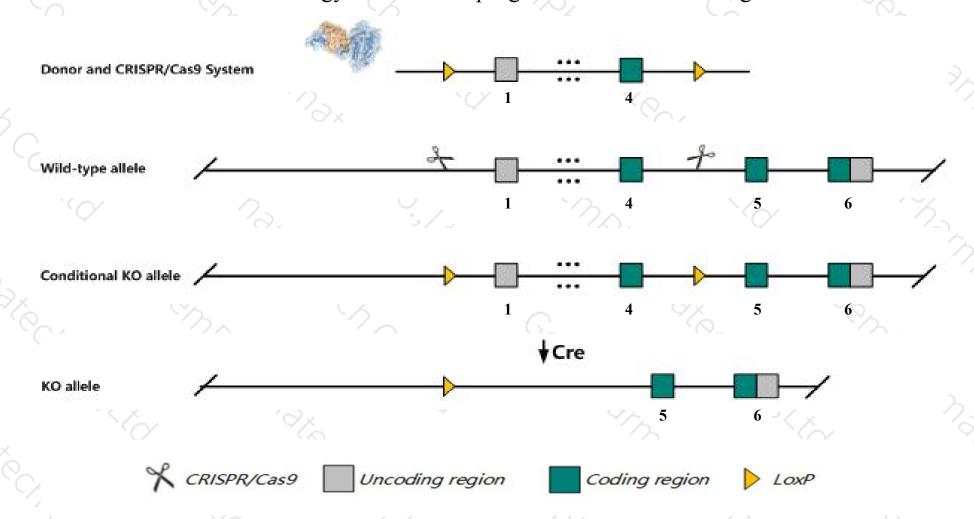
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Acp5* gene. The schematic diagram is as follows:



Technical routes



- The *Acp5* gene has 6 transcripts. According to the structure of *Acp5* gene, exon1-exon4 of *Acp5-206* (ENSMUST00000217643.1) transcript is recommended as the knockout region. The region contains start coding ATG. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Acp5* gene. The brief process is as follows:gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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,homozygous mutation of this gene results in skeletal defects such as osteopetrosis, and shortening and widening of the bones. heterozygous mutants display the same phenotype with lesser severity.
- The KO region contains the 6530413G14Rik gene. Knockout the region may affect the function of 6530413G14Rik gene.
- > The *Acp5* 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)



Acp5 acid phosphatase 5, tartrate resistant [Mus musculus (house mouse)]

Gene ID: 11433, updated on 13-Mar-2020

Summary

↑ ?

Official Symbol Acp5 provided by MGI

Official Full Name acid phosphatase 5, tartrate resistant provided by MGI

Primary source MGI:MGI:87883

See related Ensembl: ENSMUSG00000001348

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 TRACP, TRAP

Expression Broad expression in subcutaneous fat pad adult (RPKM 123.7), colon adult (RPKM 116.7) and 17 other tissuesSee more

Orthologs human all

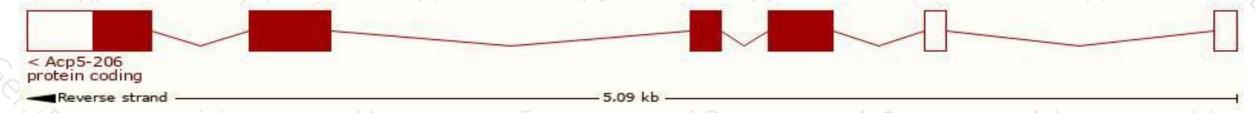
Transcript information (Ensembl)



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

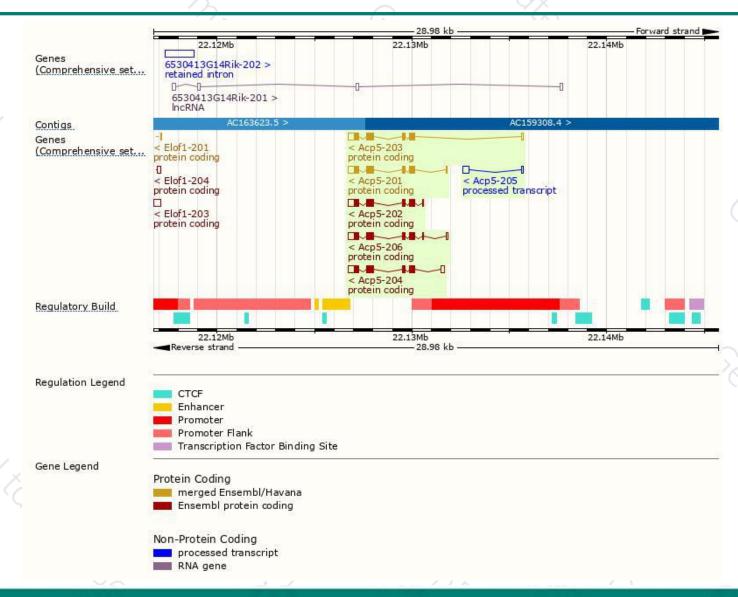
	/ 3 / mu					
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST00000217643.1	1447	<u>327aa</u>	Protein coding	CCDS22923	Q05117	TSL:5 GENCODE basic APPRIS P1
ENSMUST00000213815.1	1419	<u>327aa</u>	Protein coding	CCDS22923	Q05117	TSL:5 GENCODE basic APPRIS P1
ENSMUST00000165735.8	1376	<u>327aa</u>	Protein coding	CCDS22923	Q05117	TSL:1 GENCODE basic APPRIS P1
ENSMUST00000069330.13	1356	327aa	Protein coding	CCDS22923	Q05117	TSL:1 GENCODE basic APPRIS P1
ENSMUST00000115315.2	1349	<u>327aa</u>	Protein coding	CCDS22923	Q05117	TSL:2 GENCODE basic APPRIS P1
ENSMUST00000216684.1	399	No protein	Processed transcript	19-	2-	TSL:2
	ENSMUST00000217643.1 ENSMUST00000213815.1 ENSMUST00000165735.8 ENSMUST00000069330.13 ENSMUST000000115315.2	ENSMUST00000217643.1 1447 ENSMUST00000213815.1 1419 ENSMUST00000165735.8 1376 ENSMUST00000069330.13 1356 ENSMUST000000115315.2 1349	ENSMUST00000217643.1 1447 327aa ENSMUST00000213815.1 1419 327aa ENSMUST00000165735.8 1376 327aa ENSMUST00000069330.13 1356 327aa ENSMUST00000115315.2 1349 327aa	ENSMUST00000217643.1 1447 327aa Protein coding ENSMUST00000213815.1 1419 327aa Protein coding ENSMUST00000165735.8 1376 327aa Protein coding ENSMUST00000069330.13 1356 327aa Protein coding ENSMUST00000115315.2 1349 327aa Protein coding	ENSMUST00000217643.1 1447 327aa Protein coding CCDS22923 ENSMUST00000213815.1 1419 327aa Protein coding CCDS22923 ENSMUST00000165735.8 1376 327aa Protein coding CCDS22923 ENSMUST00000069330.13 1356 327aa Protein coding CCDS22923 ENSMUST00000115315.2 1349 327aa Protein coding CCDS22923	ENSMUST00000217643.1 1447 327aa Protein coding CCDS22923 Q05117 ENSMUST00000213815.1 1419 327aa Protein coding CCDS22923 Q05117 ENSMUST00000165735.8 1376 327aa Protein coding CCDS22923 Q05117 ENSMUST00000069330.13 1356 327aa Protein coding CCDS22923 Q05117 ENSMUST00000115315.2 1349 327aa Protein coding CCDS22923 Q05117

The strategy is based on the design of *Acp5-206* transcript, the transcription is shown below:



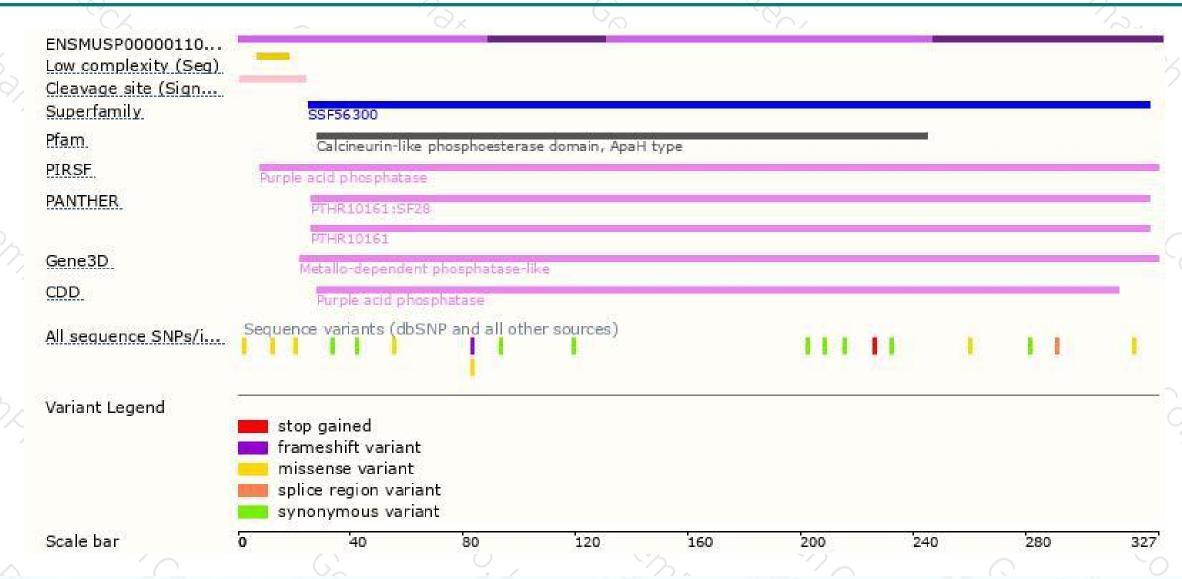
Genomic location distribution





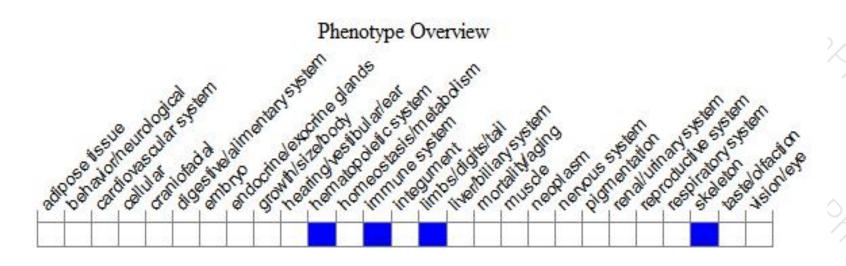
Protein domain





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, homozygous mutation of this gene results in skeletal defects such as osteopetrosis, and shortening and widening of the bones. Heterozygous mutants display the same phenotype with lesser severity.



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





