

Ankrd11 Cas9-KO Strategy

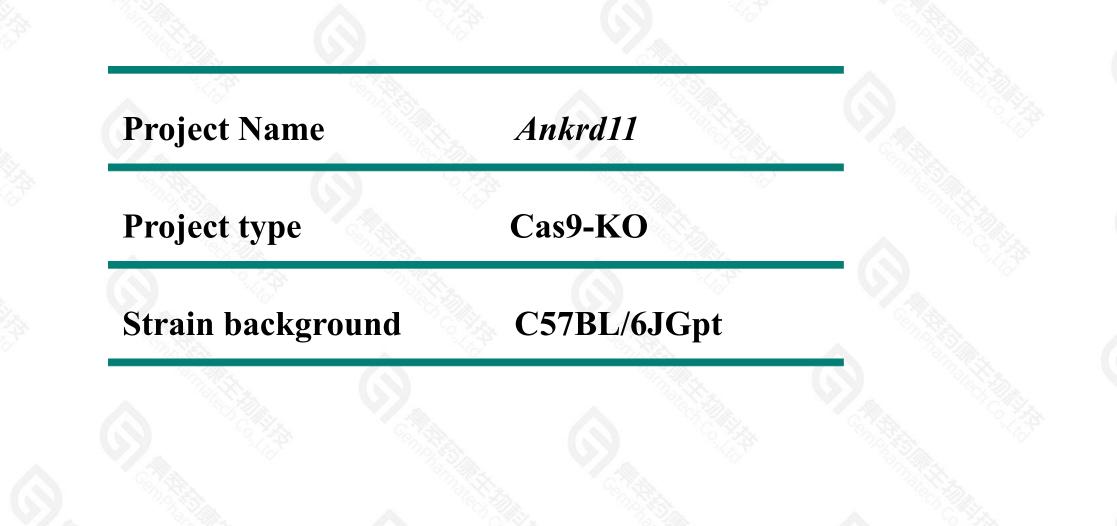
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Reviewer: Xueting Zhang

Design Date: 2021-12-17

Project Overview





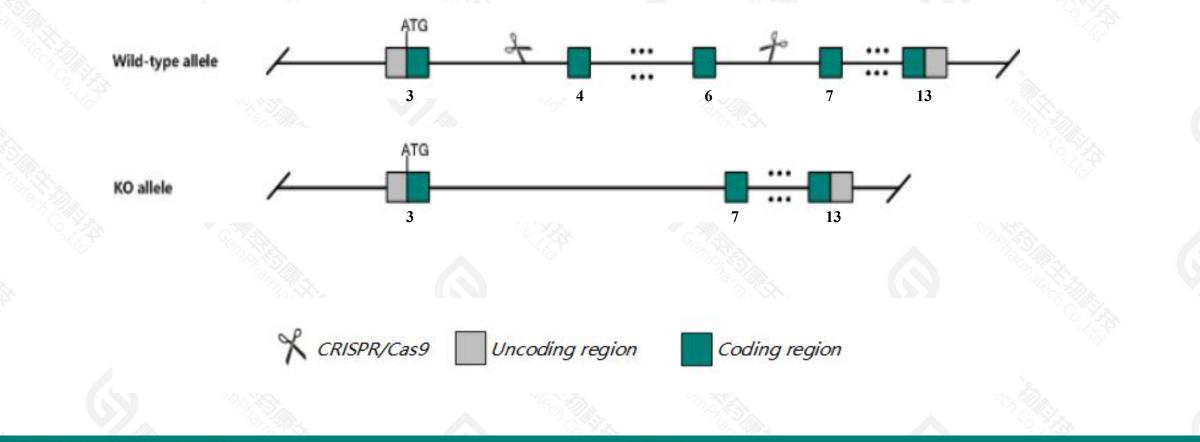
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Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ankrd11* gene. The schematic diagram is as follows:



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➤ The Ankrd11 gene has 7 transcripts. According to the structure of Ankrd11 gene, exon4-exon6 of Ankrd11-202(ENSMUST00000098334.13) transcript is recommended as the knockout region. The region contains 514bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Ankrd11* 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.



- According to the existing MGI data, mice homozygous for a spontaneous allele die by E9.5, are small and fail to turn.
 Mice heterozygous for a spontaneous allele exhibit craniofacial abnormalities, decreased weight, osteoporosis and osteopenia.
- ➤ The *Ankrd11*-207 transcript was not affected and the effect is unknown.
- > This strategy can destroy Gm45894 gene while knocking out the target gene.
- > The *Ankrd11* gene is located on the Chr8. 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)

Ankrd11 ankyrin repeat domain 11 [Mus musculus (house mouse)]

Gene ID: 77087, updated on 17-Dec-2020

Summary

Official Symbol	Ankrd11 provided by MGI
Official Full Name	ankyrin repeat domain 11 provided by <u>MGI</u>
Primary source	MGI:MGI:1924337
See related	Ensembl:ENSMUSG0000035569
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	2410104C19Rik, 3010027A04Rik, 6330578C09Rik, 9530048I21Rik, AA930108, Gm176, Yo, Yod
Expression	Ubiquitous expression in thymus adult (RPKM 25.9), ovary adult (RPKM 21.9) and 28 other tissuesSee more
Orthologs	human all

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Transcript information (Ensembl)



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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
Ankrd11-202	ENSMUST0000098334.13	8455	<u>2643aa</u>	Protein coding	CCDS40507		TSL:5, GENCODE basic, APPRIS P2,	
Ankrd11-201	ENSMUST0000098333.5	7995	<u>2664aa</u>	Protein coding	-		TSL:5 , GENCODE basic , APPRIS ALT2	
Ankrd11-205	ENSMUST00000212050.2	1874	<u>134aa</u>	Protein coding	2		CDS 5' incomplete , TSL:1 ,	
Ankrd11-207	ENSMUST00000212937.2	943	<u>121aa</u>	Protein coding	-		TSL:1 , GENCODE basic ,	
Ankrd11-203	ENSMUST00000172906.8	8481	<u>97aa</u>	Nonsense mediated decay	¥.		TSL:1,	
Ankrd11-206	ENSMUST00000212337.2	4257	No protein	Retained intron			TSL:1 ,	
Ankrd11-204	ENSMUST00000174833.2	2934	No protein	Retained intron	-		TSL:1,	

The strategy is based on the design of *Ankrd11-202* transcript, the transcription is shown below:

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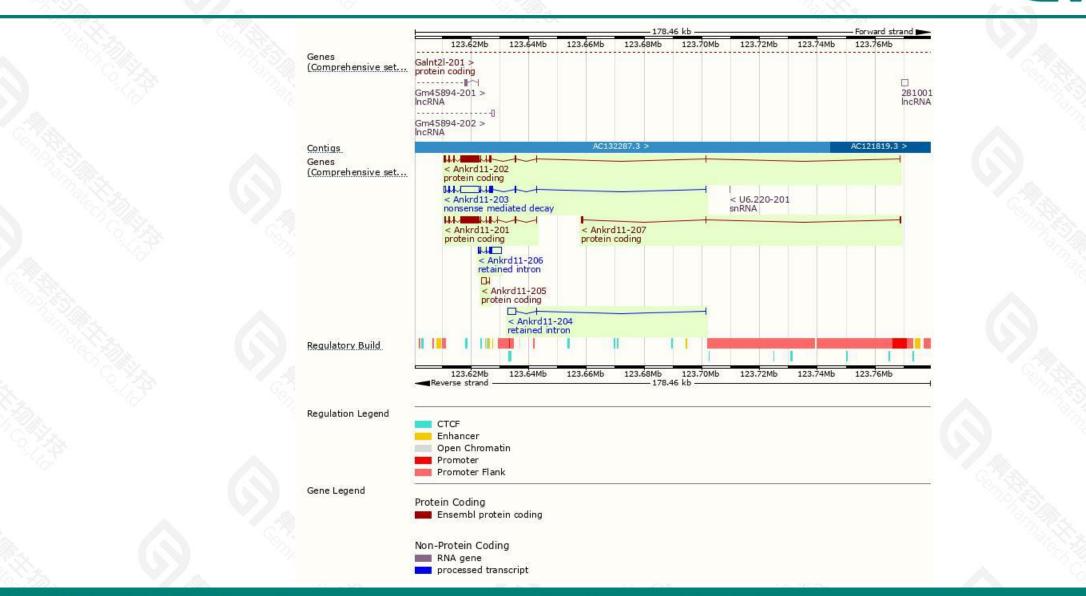
< Ankrd11-202 protein coding

Reverse strand

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158.20 kb

Genomic location distribution



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400-9660890

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Protein domain

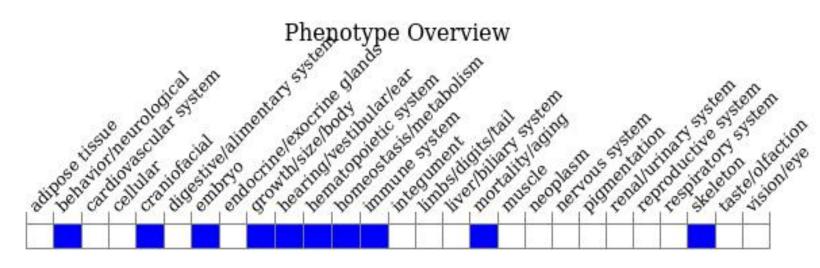


ENSMUSP00000095 MobiDB lite		_				
Low complexity (Seq)						
Coiled-coils (Ncoils)		70770 (1970) 				
Superfamily	Ankyrin repeat-containir	ig domain superfamily				
SMART	Ankyrin repeat					
<u>Pfam</u>	Ankyrin repeat Ankyrin repeat-containir	ng domain				
PROSITE profiles	Ankyrin repeat Ankyrin repeat-containir					
PANTHER	Ankyrin repeat domain-containir					
Gene3D	Ankyrin repeat-containin					
All sequence SNPs/i	Sequence variants (dbSNP a	nd all other sources)	in como por	10061-0000-000		
Variant Legend	missense variant splice region variant synonymous variant					
Scale bar	0 400	800	1200	1600	2000	2643

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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 spontaneous allele die by E9.5, are small and fail to turn. Mice heterozygous for a spontaneous allele exhibit craniofacial abnormalities, decreased weight, osteoporosis and osteopenia.

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



