

Zc3h14 Cas9-CKO Strategy

Designer: Daohua Xu

Reviewer: Huimin Su

Design Date: 2020-4-20

Project Overview



Project Name

Zc3h14

Project type

Cas9-CKO

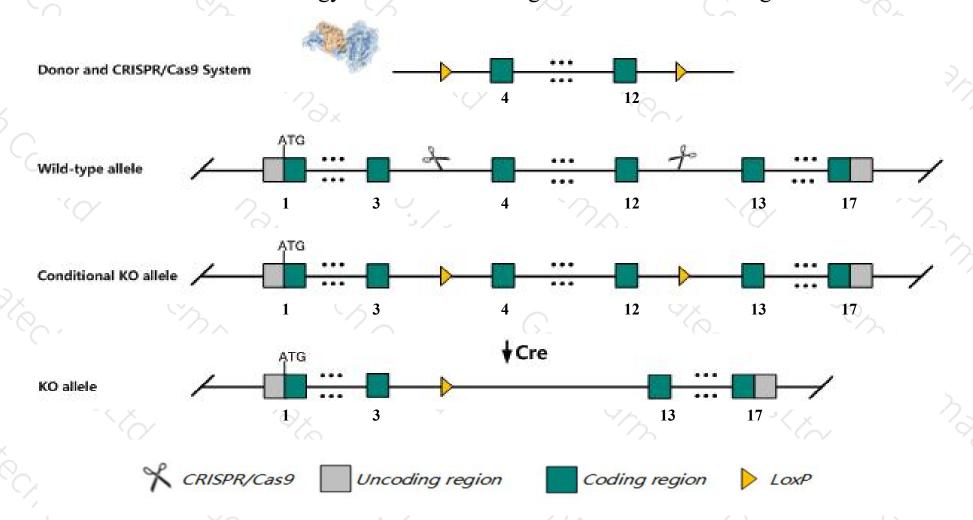
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Zc3h14 gene. The schematic diagram is as follows:



Technical routes



- The Zc3h14 gene has 13 transcripts. According to the structure of Zc3h14 gene, exon4-exon12 of Zc3h14-204 (ENSMUST00000110105.9) transcript is recommended as the knockout region. The region contains 1553bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify Zc3h14 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, homozygous knockout results in impaired spatial working memory, enlarged anterior lateral ventricles in the brain, small testes and reduced litter size.
- The Zc3h14 gene is located on the Chr12. 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)



Zc3h14 zinc finger CCCH type containing 14 [Mus musculus (house mouse)]

Gene ID: 75553, updated on 13-Mar-2020

Summary

↑ ?

Official Symbol Zc3h14 provided by MGI

Official Full Name zinc finger CCCH type containing 14 provided by MGI

Primary source MGI:MGI:1919824

See related Ensembl:ENSMUSG00000021012

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 1010001P15Rik, 1700016A15Rik, 2700069A02Rik, AU014748

Expression Ubiquitous expression in testis adult (RPKM 36.2), CNS E11.5 (RPKM 18.5) and 24 other tissuesSee more

Orthologs <u>human</u> all

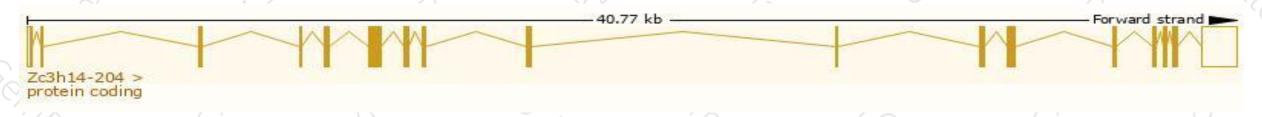
Transcript information (Ensembl)



The gene has 13 transcripts, all transcripts are shown below:

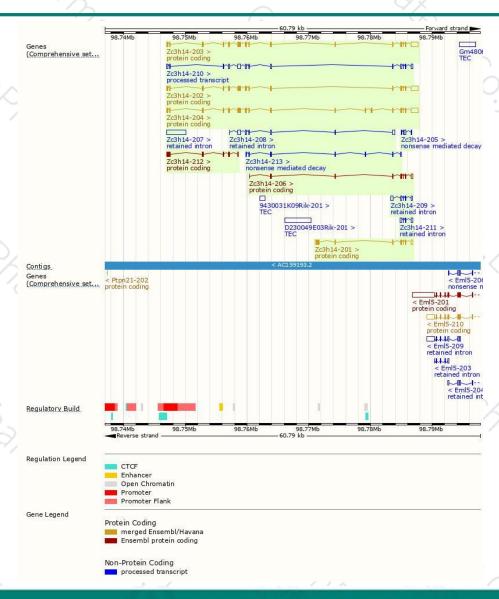
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Zc3h14-204	ENSMUST00000110105.9	3514	735aa	Protein coding	CCDS26100	Q8BJ05	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P4
Zc3h14-203	ENSMUST00000110104.9	3146	<u>604aa</u>	Protein coding	CCDS26099	Q8BJ05	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT 1
Zc3h14-202	ENSMUST00000057000.16	3046	<u>579aa</u>	Protein coding	CCDS49138	Q8BJ05	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALTZ
Zc3h14-201	ENSMUST00000021399.8	1430	309aa	Protein coding	CCDS49139	Q8BJ05	TSL:1 GENCODE basic
Zc3h14-206	ENSMUST00000221532.1	983	238aa	Protein coding	-	A0A1Y7VK20	CDS 5' incomplete TSL:5
Zc3h14-212	ENSMUST00000223083.1	924	269aa	Protein coding	(9)	A0A1Y7VK72	CDS 3' incomplete TSL'2
Zc3h14-213	ENSMUST00000223451.1	727	<u>180aa</u>	Nonsense mediated decay	V/40	A0A1Y7VJD9	CDS 5' incomplete TSL:5
Zc3h14-205	ENSMUST00000220660.1	598	70aa	Nonsense mediated decay		A0A1Y7VP61	CDS 5' incomplete TSL:5
Zc3h14-210	ENSMUST00000222461.1	2012	No protein	Processed transcript	(5)	8	TSL:1
Zc3h14-207	ENSMUST00000221558.1	3078	No protein	Retained intron	(9)		TSL:NA
Zc3h14-208	ENSMUST00000221576.1	1351	No protein	Retained intron	V20	ū.	TSL:1
Zc3h14-209	ENSMUST00000222146.1	895	No protein	Retained intron		2	TSL:2
Zc3h14-211	ENSMUST00000222632.1	687	No protein	Retained intron	-		TSL:2

The strategy is based on the design of Zc3h14-204 transcript, the transcription is shown below:



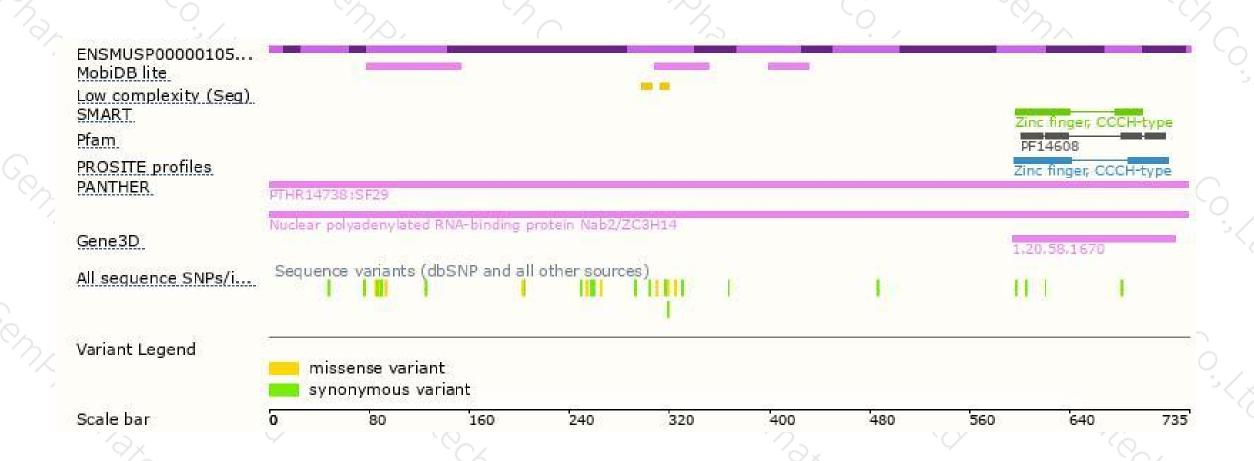
Genomic location distribution





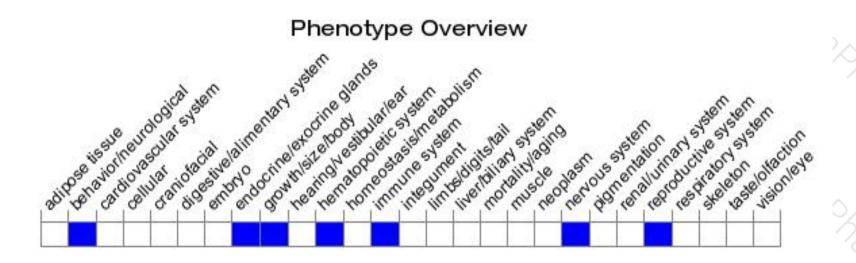
Protein domain





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 knockout results in impaired spatial working memory, enlarged anterior lateral ventricles in the brain, small testes and reduced litter size.



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





