Ywhab Cas9-CKO Strategy

Designer: Daohua Xu

Design Date: 2019-7-18

Project Overview



Project Name

Project type Cas9-CKO

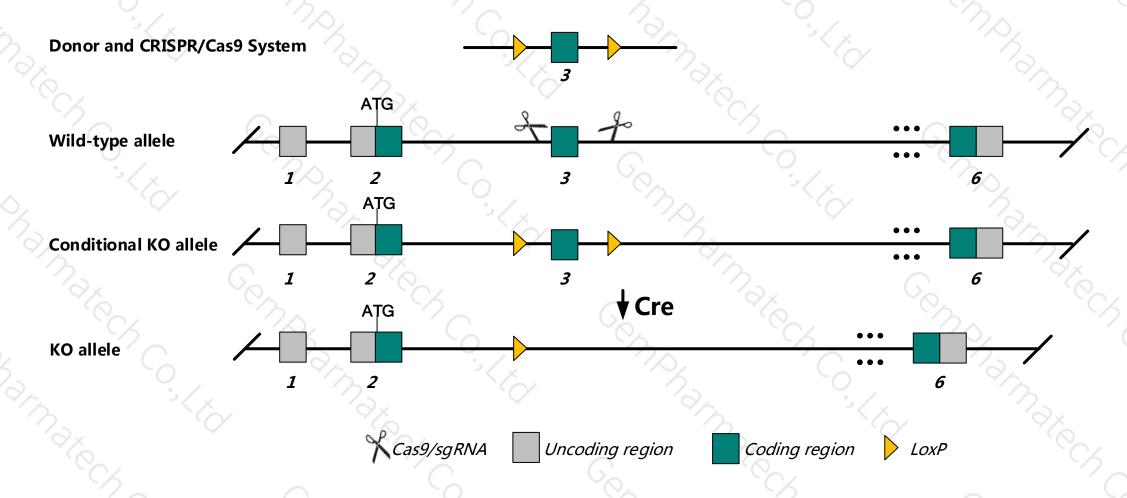
Strain background C57BL/6JGpt

Ywhab

Conditional Knockout strategy



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



Technical routes



- The *Ywhab* gene has 2 transcripts. According to the structure of *Ywhab* gene, exon3 of *Ywhab*-201 (ENSMUST00000018470.9) transcript is recommended as the knockout region. The region contains 124bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ywhab* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.

Notice



- The *Ywhab* gene is located on the Chr2. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)



Ywhab tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, beta polypeptide [Mus musculus (house mouse)]

Gene ID: 54401, updated on 9-Dec-2018

Summary

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Official Symbol Ywhab provided by MGI

Official Full Name tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, beta polypeptide provided by MGI

Primary source MGI:MGI:1891917

See related Ensembl: ENSMUSG00000018326

Gene type protein coding RefSeq status PROVISIONAL Organism Mus musculus

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

Muroidea; Muridae; Murinae; Mus; Mus

Also known as 14-3-3b; 1300003C17Rik

Expression Ubiquitous expression in cortex adult (RPKM 73.1), CNS E18 (RPKM 72.1) and 28 other tissues See more

Orthologs human all

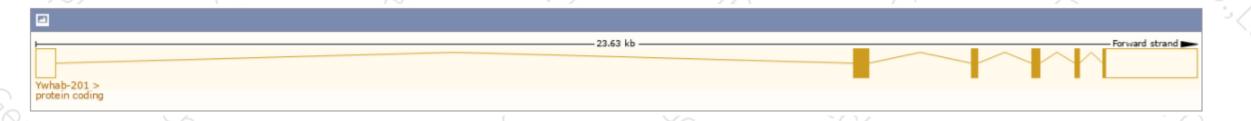
Transcript information (Ensembl)



The gene has 2 transcripts, and all transcripts are shown below:

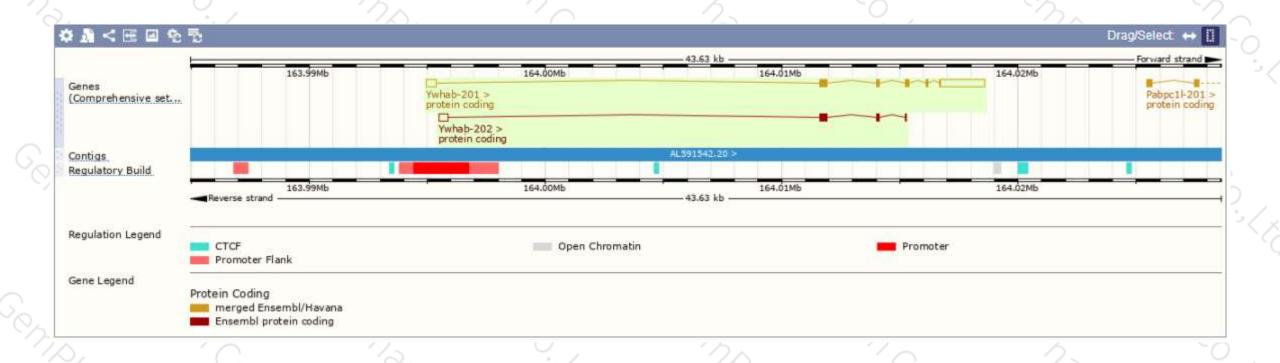
Show/hide columns (1 hidden)							
Name 🍦	Transcript ID 🗼	bp Prote	tein 🍦 Biotype 👙	CCDS	UniProt	RefSeq	Flags 🝦
Ywhab-201	ENSMUST00000018470.9	3013 246	Protein coding	CCDS17019₽	A2A5N2@Q9CQV8@	NM 018753 ଜ NP 061223 ଜ	TSL:1 GENCODE basic APPRIS P1
Ywhab-202	ENSMUST00000131288.1	849 <u>159</u>	Protein coding	-	<u>A2A5N1</u> @	-	CDS 3' incomplete TSL:3

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



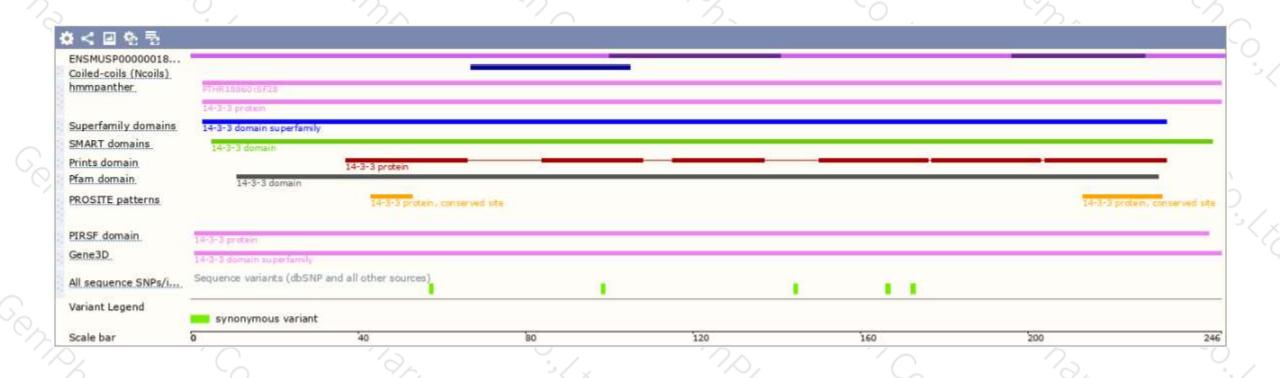
Genomic location distribution





Protein domain





If you have any questions, you are welcome to inquire. Tel: 025-5864 1534





