

# Epha2 Cas9-CKO Strategy

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**Reviewer:** 

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# **Project Overview**



**Project Name** 

Epha2

**Project type** 

Cas9-CKO

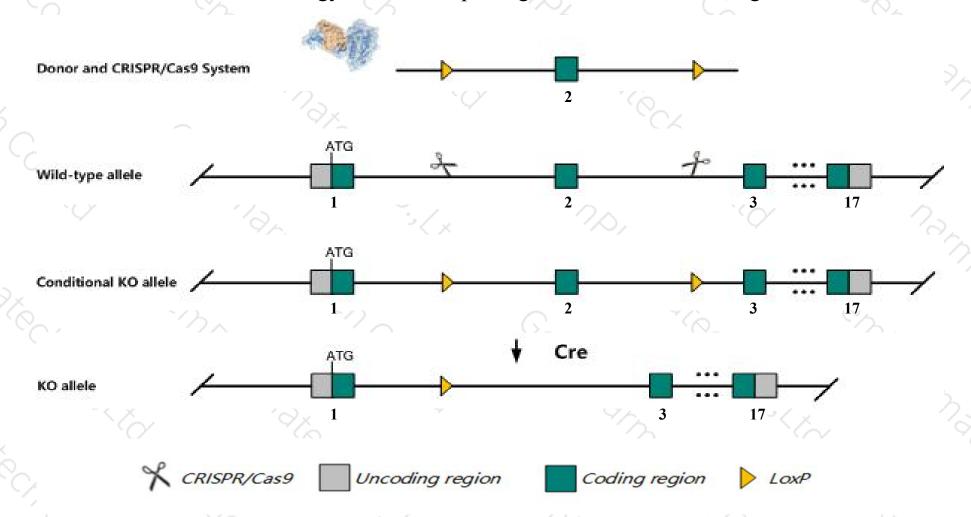
Strain background

C57BL/6JGpt

# Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Epha2* gene. The schematic diagram is as follows:



### Technical routes



- The *Epha2* gene has 4 transcripts. According to the structure of *Epha2* gene, exon2 of *Epha2-201* (ENSMUST0000006614.2) transcript is recommended as the knockout region. The region contains 68bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Epha2* 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, Mice homozygous for a null allele exhibit abnormal angiogenesis. Mice homozygous for a gene trap allele exhibit increased incidence of chemically-induced tumors, increased metastatic potential, and age-related cataracts.
- > The *Epha2* gene is located on the Chr4. 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)



#### Epha2 Eph receptor A2 [Mus musculus (house mouse)]

Gene ID: 13836, updated on 19-Feb-2019

#### Summary

☆ ?

Official Symbol Epha2 provided by MGI

Official Full Name Eph receptor A2 provided by MGI

Primary source MGI:MGI:95278

See related Ensembl:ENSMUSG00000006445

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 AW545284, Eck, Myk2, Sek-2, Sek2

Expression Broad expression in lung adult (RPKM 25.3), small intestine adult (RPKM 22.7) and 19 other tissuesSee more

Orthologs <u>human</u> all

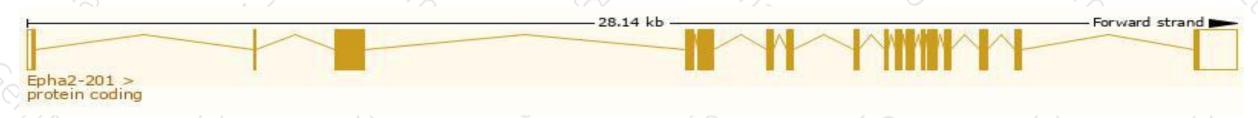
# Transcript information (Ensembl)



The gene has 4 transcripts, all transcripts are shown below:

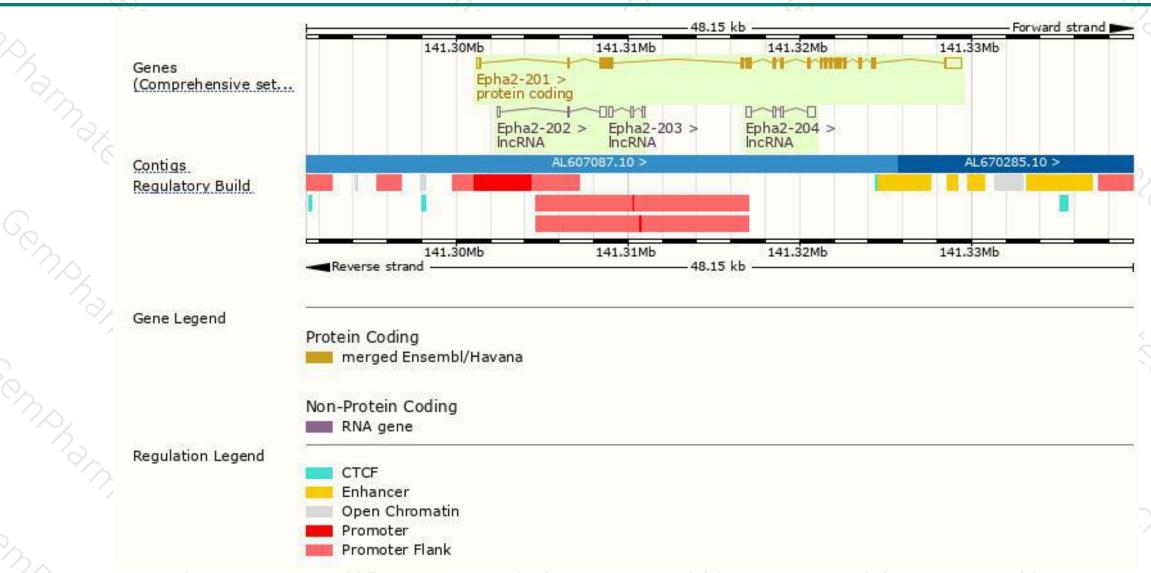
Name	Transcript ID	bp	Protein	Biotype	ccds	UniProt	Flags
Epha2-201	ENSMUST00000006614.2	3913	<u>977aa</u>	Protein coding	CCDS18869	Q03145	TSL:1 GENCODE basic APPRIS P1
Epha2-204	ENSMUST00000149002.1	969	No protein	IncRNA	-	35.3	TSL:5
Epha2-203	ENSMUST00000145523.1	531	No protein	IncRNA	ų.	925	TSL:3
Epha2-202	ENSMUST00000131026.1	507	No protein	IncRNA	2	727	TSL:3

The strategy is based on the design of *Epha2-201* transcript, The transcription is shown below



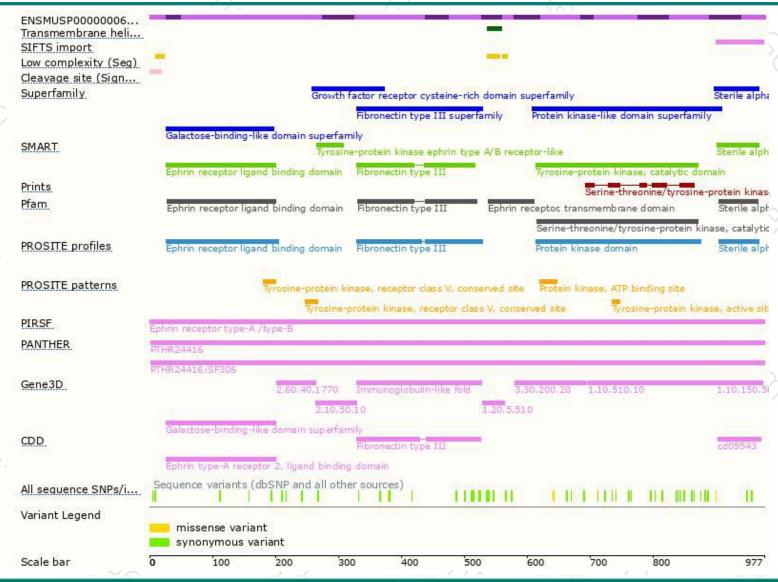
### Genomic location distribution





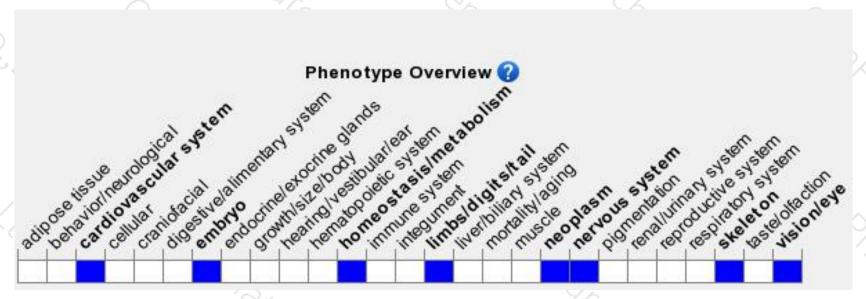
### Protein domain





# 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 null allele exhibit abnormal angiogenesis. Mice homozygous for a gene trap allele exhibit increased incidence of chemically-induced tumors, increased metastatic potential, and age-related cataracts.



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





